Evolution 60
Local Control User's Guide

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For technical assistance, please contact:
Technical Support
Thermo Fisher Scientific
5225 Verona Road
Madison WI 53711-4495
U.S.A.

Telephone: 1 800 642 6538 (U.S.A.) or +1 608 273 5017 (worldwide)
Fax: +1 608 273 5045 (worldwide)
E-mail: techsupport.analyze@thermo.com
www:thermo.com

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Introduction

Congratulations on your purchase of a Thermo Scientific Evolution 60 spectrophotometer! Our spectrophotometers integrate advanced hardware features with the power and flexibility of a wide range of accessories.

Conventions used in this manual

This manual includes safety precautions and other important information presented in the following format:

Note Notes contain helpful supplementary information. ▲

Notice Follow instructions labeled "Notice" to avoid damaging the system hardware or losing data. ▲

⚠ Caution Indicates a hazardous situation which, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices. ▲

⚠ Warning Indicates a hazardous situation which, if not avoided, could result in death or serious injury. ▲

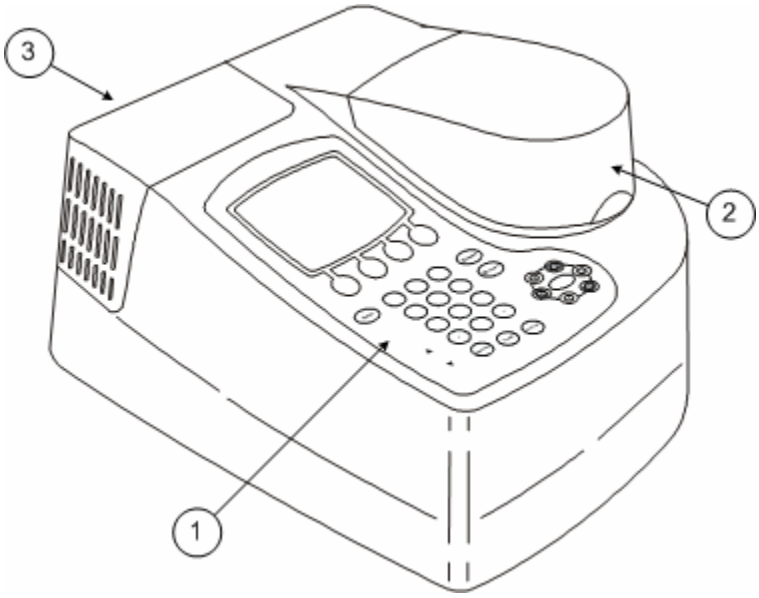
⚠ Danger Indicates a hazardous situation which, if not avoided, will result in death or serious injury. ▲

Spectrophotometer Basics

This chapter describes the major components of your spectrophotometer.

Spectrophotometer components

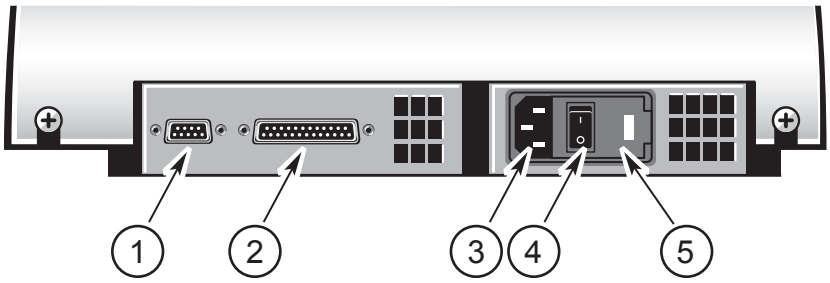
The following illustration identifies some major components visible on the outside of a typical Evolution 60 spectrophotometer.



Key	
①	Keypad (not present on PC only models)
②	Sample Compartment
③	Optional printer housing

Connectors

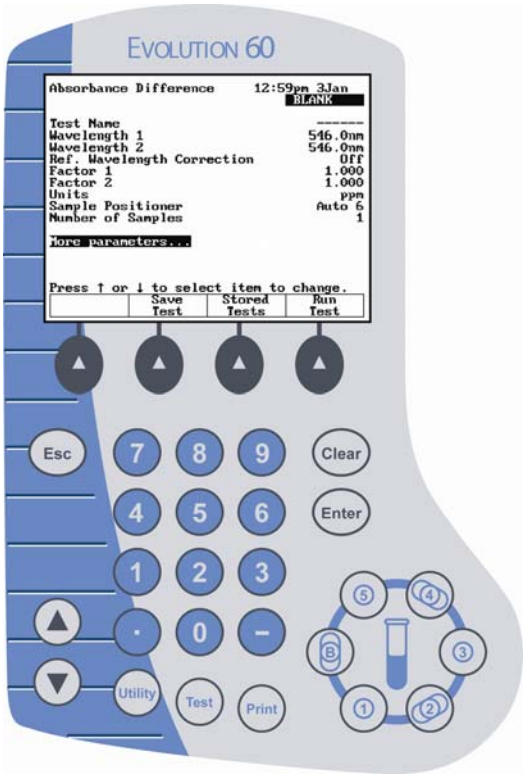
The following illustrations show the locations of the connectors on the back of the spectrophotometer.



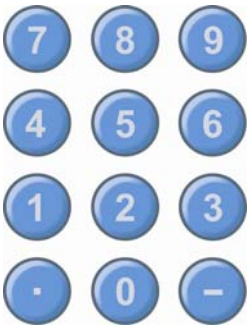
Key	
①	RS232C port
②	Parallel printer port
③	A/C power connector
④	On/Off switch
⑤	Fuse compartment

⚠ Warning Avoid shock hazard. Always turn off the instrument and unplug it from the wall outlet or power strip before you unplug the power cord from the instrument connector. ▲

About the keypad



Key or Button	Function
	• Called 'Function' keys • Allows you to perform a specific function displayed on the screen above the appropriate key • Functions change from one screen to another • Depending on the application, some function keys will not be active
	• Can clear value being entered • Does not change any values you have already accepted • May return to the previous screen
	• Deletes last character entered • Does not change any values you have already accepted • Does not return to the previous screen

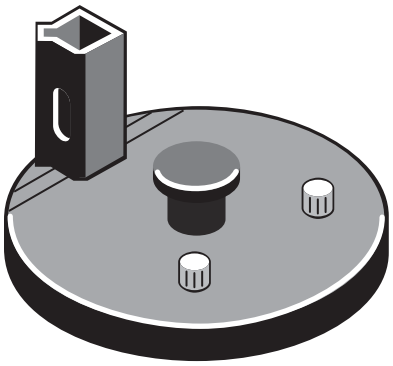
Key or Button	Function
	• Accepts highlighted or selected values • Advances to next parameter or screen
	• Prints the displayed information
	• Displays the Test Types screen
	• Displays the Utility Screen
	• Controls the location of the cursor • Highlights the value or option for selection
	• Enters numbers, decimal point and minus sign for entry of values
	• Cell position keys • Selects cell holder position to be measured • B = blank and 1-5 = sample positions • = positions when used in Auto 3 mode

Cell Holders

All models of the Evolution 60 include a 6-position and single cell holder.

6-Position Cell Holder

Single Cell Holder



Note Whenever you press *Run Test* in any of the test modes to start a measurement, the instrument will attempt to initialize the cell positioner. If a single cell holder is installed, the message "Error, Single Cell Holder found. Use Single Cell Holder?" is displayed. Press *Accept Change* to continue, or install the cell changer and press *Cancel Change*. ▲

For a more detailed list of accessories available, please see the parts list.

Selecting and positioning cuvettes

The compatible wavelength range for different types of cells depends upon the material used.

Cell Type	Wavelength
Glass:	
Optical Glass	360 to > 1100 nm
Special Optical Glass	320 to > 1100 nm
Pyrex	320 to > 1100 nm
Borosilicate Glass	330 to > 1100 nm
Quartz (also called silica):	
Herasil Quartz	230 to > 1100 nm
Infrasil (IR Quartz)	220 to > 1100 nm
Suprasil	200 to > 1100 nm
ES Quartz	190 to > 1100 nm
Spectrosil (Far UV)	170 to > 1100 nm
Disposable:	
Polystyrene	> 340 nm
Methacrylate	> 300 nm
Acrylic	> 280 nm
UV-transparent Plastic	> 220 nm
Refer to manufacturer's specifications and ensure that you work within the recommended range.	

Test tubes vs. cuvettes

Square cuvettes yield very precise results. However, matched test tubes, when properly handled, can show as little as 1-2% deviation between readings.

The pathlength of test tubes is not as well defined as in square cuvettes. However, constructing a standard curve eliminates the need for great accuracy in knowing the pathlength, provided that the same pathlength cell is used for all blanks, standards and samples.

Other guidelines

Be sure to position cuvettes and test tubes so that the clear sides face the light beam. This means that one clear side should face the front of the instrument and the other clear side should face the back of the instrument.

Note Test tubes should always be placed in the instrument in exactly the same orientation in the light beam. An alignment mark on the test tube helps you orient the test tubes consistently and correctly. ▲

When using small aperture cells:

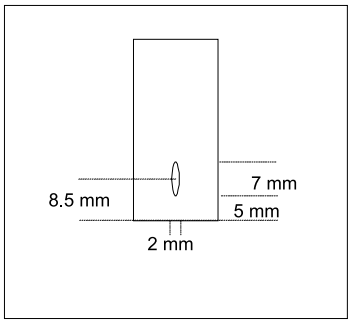
- Always used masked cells
- Use the same cell (or cuvette) for your blank and your samples

Z-dimensions

The figure below illustrates the position of the light beam in the spectrophotometer.

The specifications for the compartment, including the dimensions of the beam are:

- Z-dimension
  - Square cuvettes/test tubes 8.5 mm
  - Beam size: 2 mm (wide) by 7 mm (high)

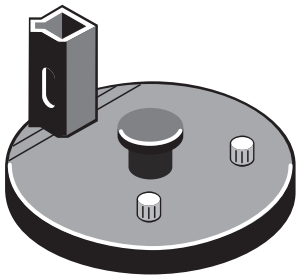


Cell holder configurations

The following table shows the cell holders and cell holder accessories available for the spectrophotometer.

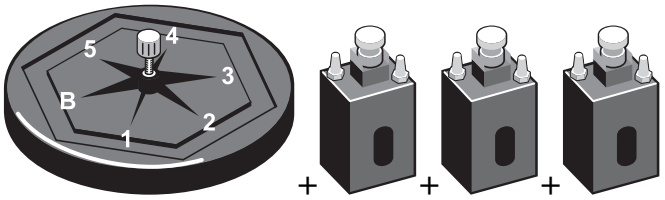
These accessories may be installed or removed without the need to switch off the instrument.

Standard Cell Holders

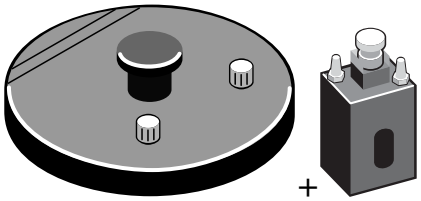


Accessory Cell Holder Systems

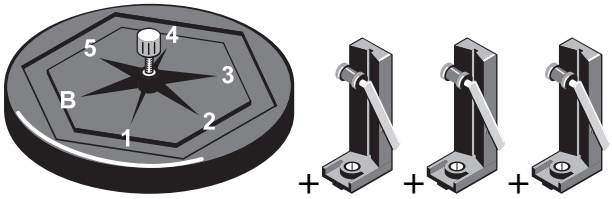
Single Cell Recirculator Temperature Control



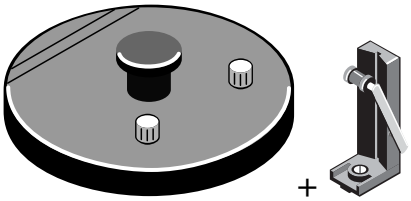
Must be installed in the B, 2 and 4 positions



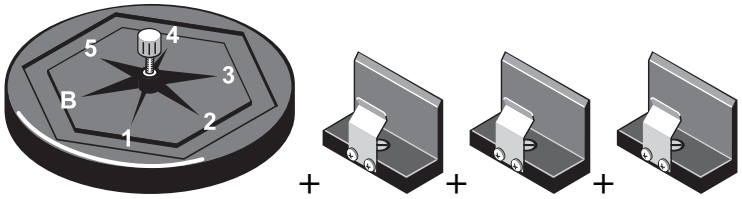
Test Tube Holder



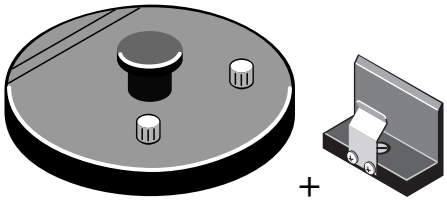
Must be installed in the B, 2 and 4 positions



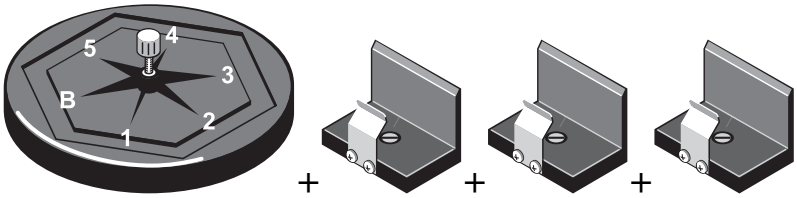
50 mm Rectangular Long Pathlength Cell Holder



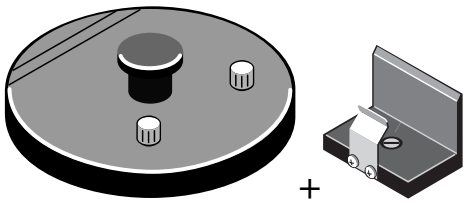
Must be installed in the B, 2 and 4 positions



50 mm Cylindrical Long Pathlength Cell Holder



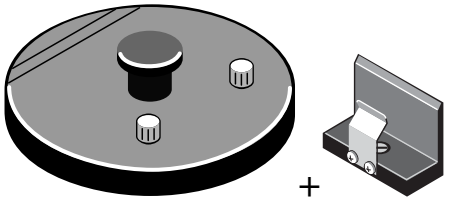
Must be installed in the B, 2 and 4 positions



Cell Changer System	Single Cell System

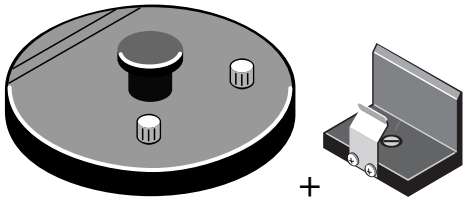
100 mm Rectangular Long Pathlength Cell Holder

Cannot use 100 mm path cells with Cell Positioner

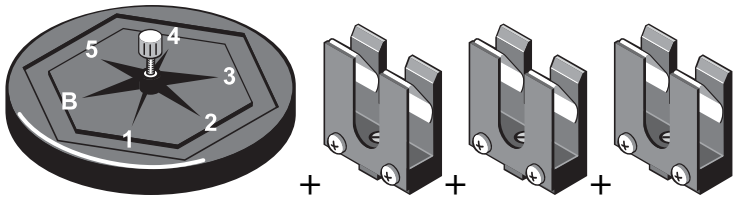


100 mm Cylindrical Long Pathlength Cell Holder

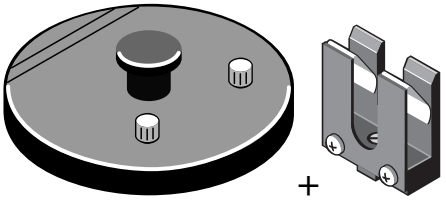
Cannot use 100 mm path cells with Cell Positioner



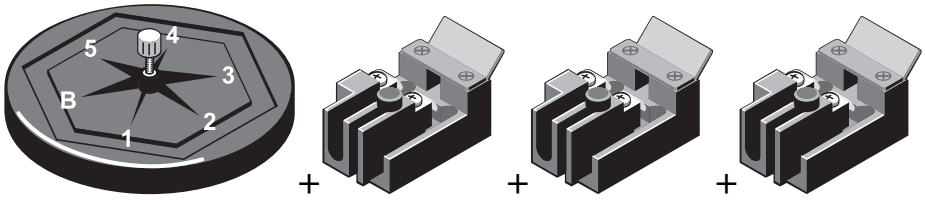
Thin Film / Filter Holders



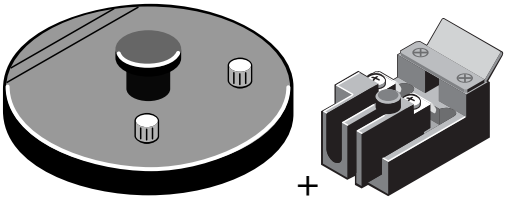
Must be installed in the B, 2 and 4 positions



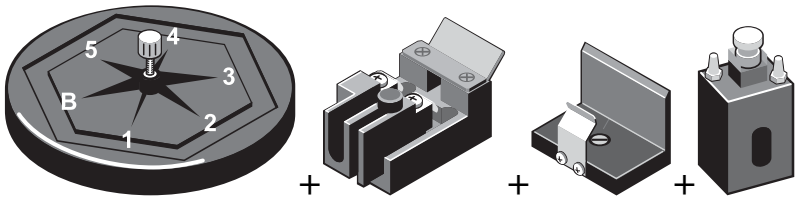
Adjustable Filter / Lens Holder



Must be installed in the B, 2 and 4 positions



Combination Systems



Must be installed in the B, 2 and 4 positions

Not applicable

Accessories

This chapter briefly describes the types of sampling and system accessories that are available for your spectrophotometer. Complete descriptions and operating instructions are included with the accessories.

These accessories may be installed or removed without the need to switch off the instrument.

Term	Description
Cell holder assembly, Cell holder accessory	Complete unit that installs in the sample compartment
Cell holder	Part that actually holds the cell in place and is installed on a baseplate
Baseplate	Part that holds the cell holder and is secured in the sample compartment floor

Cell holders and cell holder accessories

The spectrophotometer is shipped with both the 6-Position Cell Holder (installed at the factory) and the Single Cell Holder. Directions to remove and install these cell holders and other cell holder accessories are below.

Cell holder initialization

When you press Run Test to start a measurement, the instrument will display the message, "Calibrating and Checking Turret, Please Wait" while it attempts to initialize the cell changer at the Blank position.

If a single cell holder has been installed, the message "Error, Single Cell Holder found. Use Single Cell Holder?" is displayed. Press Accept Change to continue, or install the cell changer and press Cancel Change.

If the 6-Position Cell Holder has been installed, it will initialize it to the 'B' position. After it has initialized the cell changer, the instrument will display the data collection screen for the test.

Note If, while in this screen, you remove the cell changer and press Run Sample, the instrument will display "Fatal Error: 8 Press Esc to return to main menu."

Pressing *Esc* returns you to the parameter menu screen of the test. Press *Run Test* and the instrument will now display the message, "Calibrating and Checking Turret, Please Wait" while it attempts to initialize the cell positioner at the Blank position.

If the cell changer is still not in the sample compartment, the instrument will display "Error, Single Cell Holder Found. Use Single Cell Holder?"

Either press *Cancel Change* and reinstall the cell changer or press *Accept Change* to continue with using the single cell holder.

To prevent the Fatal Error whenever removing the cell changer, always return to either the main menu or the test parameter menu of the test before removing the cell changer. ▲

Changing cell holders

To:

- use long pathlength cells (cylindrical or rectangular)
- use test tubes
- measure solid samples in the filter holder
- regulate sample temperature via an external liquid recirculator

you must install the appropriate cell holders. The 6-Position Cell Holder installed in the spectrophotometer can easily be removed to install other accessory cell holders. See Removing the 6-Position Cell Holder and the Single Cell Holder.

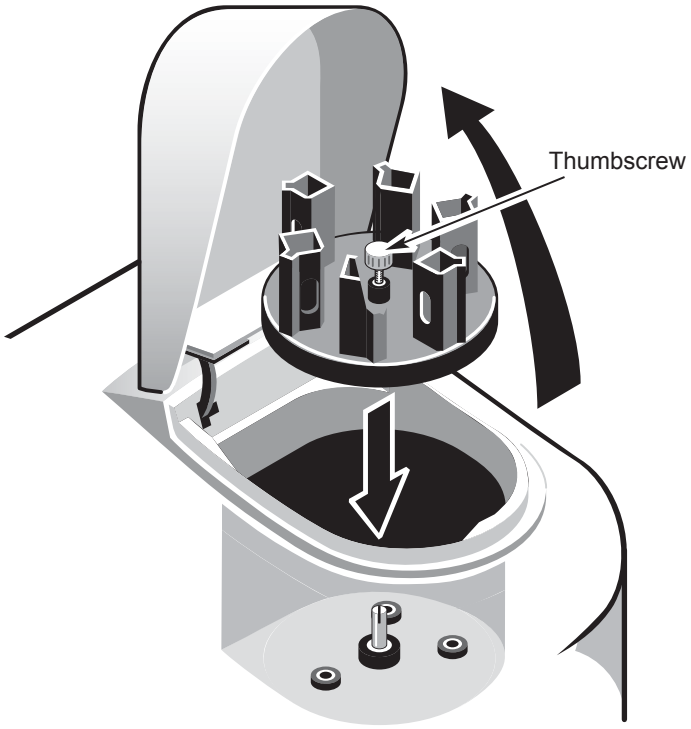
Installing the 6-Position Cell Holder and the Single Cell Holder

Follow these steps to properly install the 6-Position Cell Holder or the Single Cell Holder.

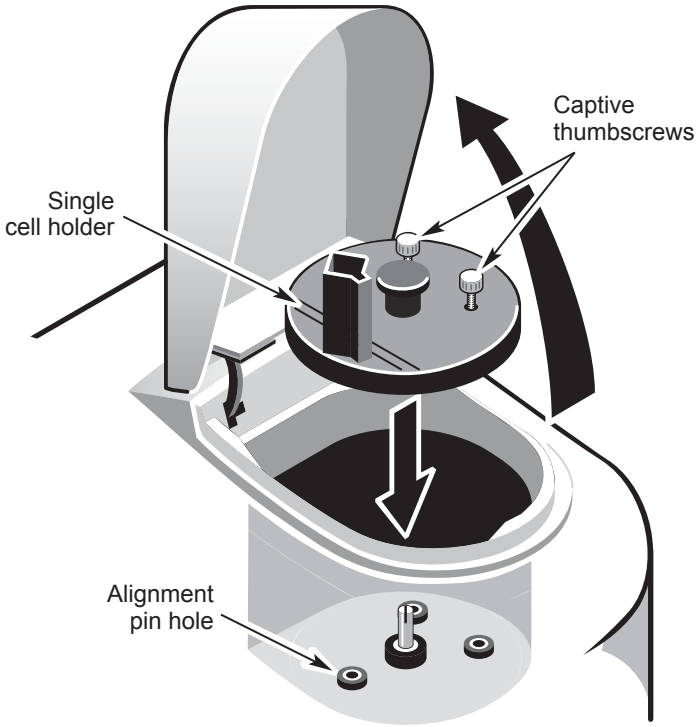
1. **Open the sample compartment door and let it rest on its hinge.**

2. **With one hand, carefully lower the cell holder straight down into the sample compartment.**

6-Position Cell Holder



Single Cell Holder



3. **With the other hand, tighten the captive thumbscrew(s).**

4. **Close the sample compartment door.**

Note If the cell holder is not aligned correctly, you will not be able to tighten the thumbscrews. ▲

Removing the 6-Position Cell Holder and the Single Cell Holder

Follow these steps to remove the 6-Position Cell Holder or the Single Cell Holder.

1. **Open the sample compartment door and let it rest on its hinge.**

2. **With one hand, loosen the captive thumbscrew.**

3. **With the other hand, pull straight up on the cell holder and lift it out of the sample compartment**

4. **Close the sample compartment door.**

Installing accessory cell holders

Make sure that you have the correct cell holder baseplate installed.

Note If you want to use 100 mm long pathlength cells, you must install the Single Cell Holder baseplate. ▲

See the Cell Holder Configurations section for the different cell holder accessories that can be created for your instrument.

Each of the Cell Holders need to be installed on either a single-cell baseplate or a multi-cell baseplate by removing the cell holder(s) secured to the baseplate. Each cell holder has a captive screw in the bottom of the holder that secures the holder to the baseplate. Use a flat blade screwdriver to loosen the captive screw from the baseplate and lift the cell holder from the baseplate. Then insert the new cell holder into the appropriate position and secure it to the baseplate by tightening the captive screw.

Note You can install only three each of some accessory cell holders. Make sure to place them in position B, 2 and 4. ▲

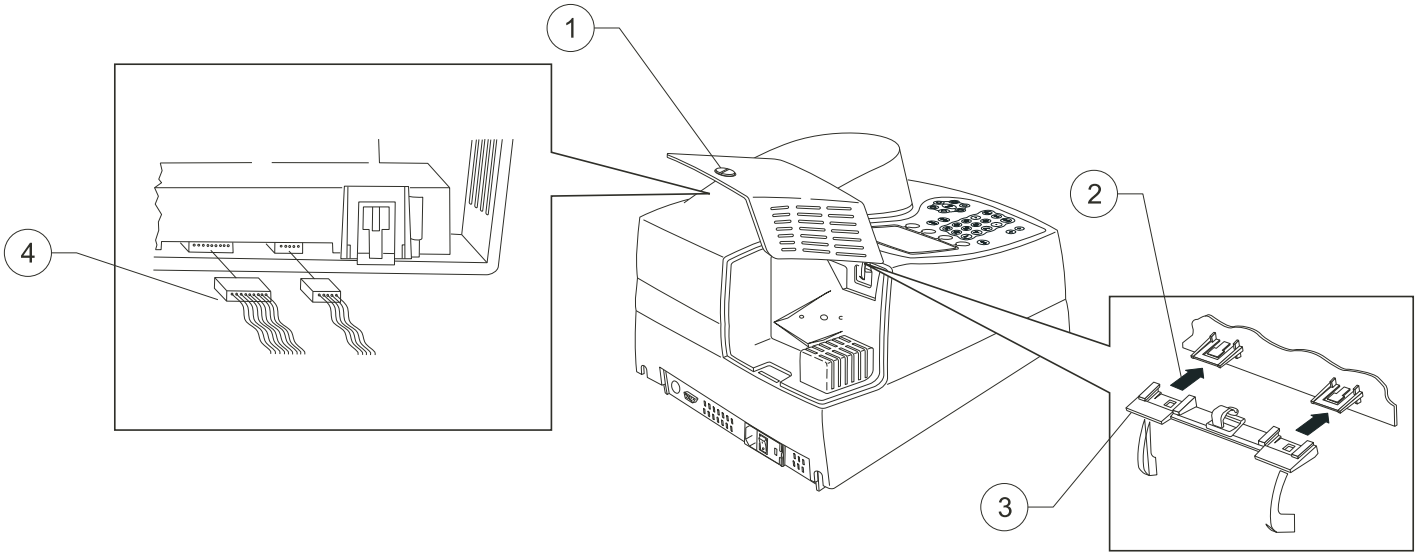
Follow the steps in the "Installing the 6-Position Cell Holder and the Single Cell Holder" section above for instructions on installing the complete accessory assembly in your instrument.

Note You can install only three each of some accessory cell holders. Make sure to place them in position B, 2 and 4. ▲

Installing the internal printer (optional)

Follow the steps below to install the optional internal printer.

⚠ Caution Turn off the instrument and disconnect the power cord from the outlet before installing the internal printer. ▲



Key	
①	Captive screw on lamp door
②	Removing door from hinge
③	Hinge
④	Connecting wires to the printer

1. **Loosen the captive screw (①) on the lamp door by rotating it counterclockwise about ¼ turn.**
2. **Open the lamp door.**
3. **Use a pen or screwdriver to lift the tabs holding the door to the hinge (② and ③).**
4. **Slide the door off the hinge.**
5. **Remove the printer (already installed on the printer door) from its packing.**
6. **Unclip the connector wires on the door hinge.**
7. **Lower the hinge so it is out of the way.**
8. **Connect the wires (④) and press into place with a small screwdriver.**

There is only one way that the connectors will fit. Each connector has a slight D shape. Make sure the side of the connector with the shiny metal contacts faces away from the printer and towards the plastic door.

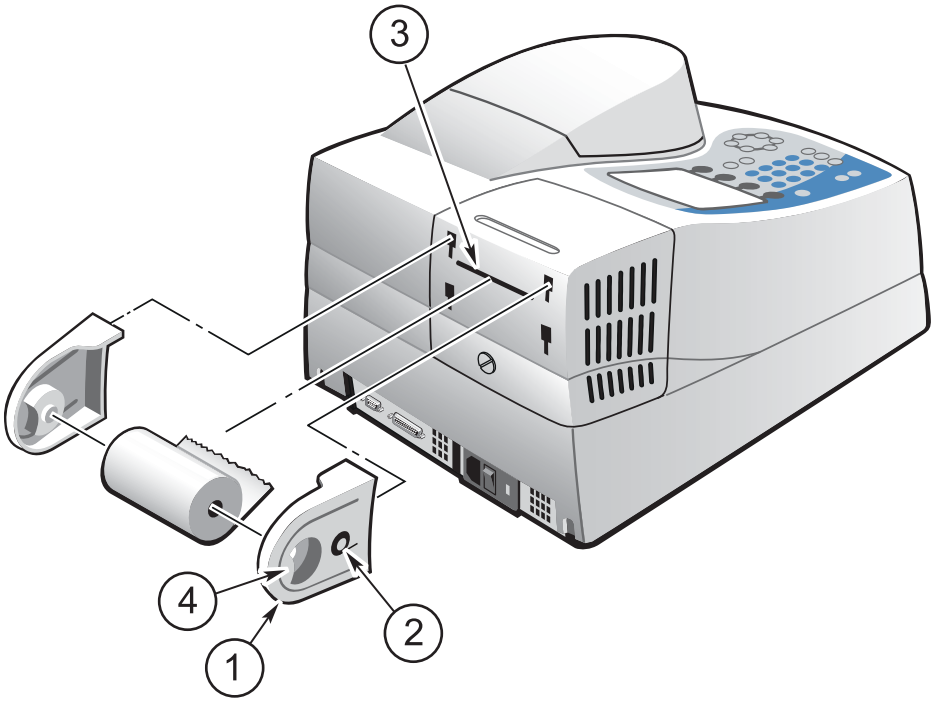
9. **Use the clip on the hinge to secure the wires.**

10. **Install the printer door by sliding it back onto the hinge (③).**

11. **Close the lamp door.**

12. **Tighten the captive screw (①) on the printer door to hold it securely in place.**

Loading paper in the internal printer



Key	
①	Paper roll holder
②	Icon for paper direction
③	Paper entry slot
④	Finger tab

Note Make sure that the paper roll holders are in place as shown. When installed correctly, they will fit flush with the top of the instrument. ▲

13. **Cut the paper so the edge is even.**

Note Arrows on the paper roll holders indicate the direction of the paper feed. ▲

14. **Feed the paper straight into the paper entry slot.**

    The printer grabs the end of the paper and pulls it in.

15. **In Basic ATC mode, when the paper stops, press *Enter* to continue advancing the paper until the paper comes out of the paper exit slot.**

16. **Pull out on the finger tabs (④) on the paper roll holders and secure the roll of paper onto the paper roll holder.**

External printers

Your spectrophotometer is able to print to external desktop printers supporting HP PCL 5.0 format and later.

Note PCL format does not support HP "Windows" printers. ▲

To print to an HP PCL printer, connect the parallel interface cable to the printer and to the parallel port on the back of the instrument (Connectors section, ②).

Note The instrument is compatible with specific printers and printer cables we offer. Contact technical support or your sales representative for more information. ▲

Note If the printer/cable is not purchased from us, it is the responsibility of the customer to determine if the printer/cable is compatible with the instrument. ▲

Setting up the Instrument

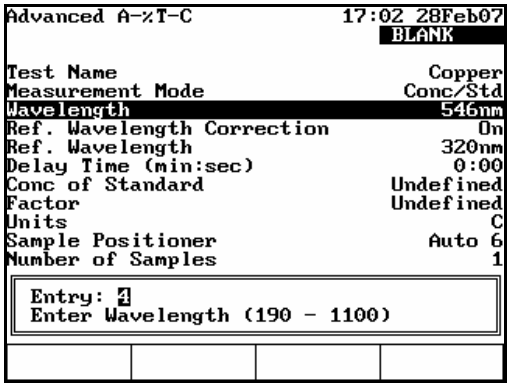
This chapter provides important information about using your instrument to analyze samples.

Entering parameter values

For models with Local Control, you will use the keypad to enter or select values. This section describes the various modes of entering or selecting parameter values.

Numeric entry

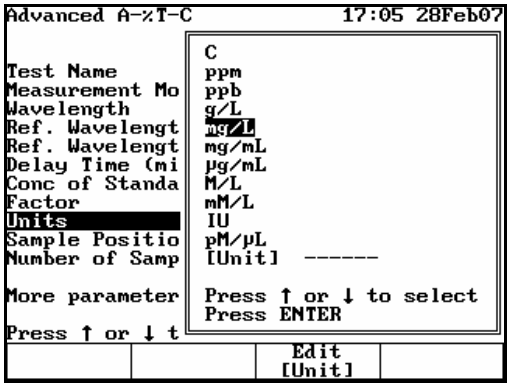
With the parameter (e.g., Wavelength) highlighted, start typing the numeric value. An Entry window with the value range appears. Type the complete entry and press *Enter*



Alternatively, you can press *Enter* to display the Entry window with the value range and then type the complete entry and press *Enter*.

Menu selection

With the parameter (e.g., Units or Sample Positioner) highlighted, press *Enter*. The selection list will appear. Highlight the appropriate item and press *Enter*.

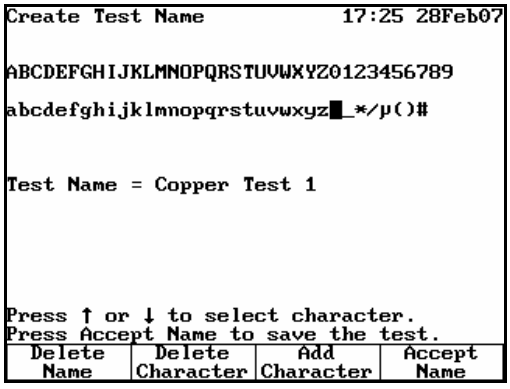


On/Off toggle

With the parameter (e.g., AutoPrint) highlighted, press *Enter*. The value will toggle to the opposite value.

Alphanumeric entry

With the parameter (e.g., Test Name) highlighted, press *Enter*. The Name Entry screen is displayed. Highlight the desired character, and press *Add Character*. When completed, press *Accept Name*.

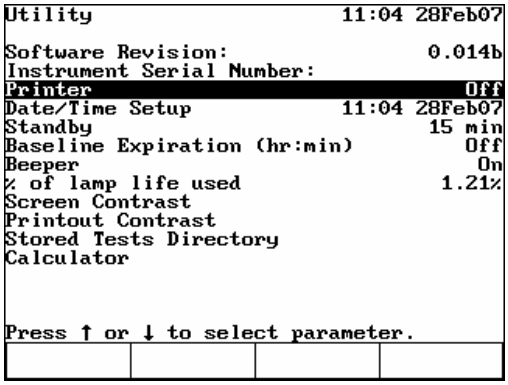


Setting utility parameters

Utility is used to set certain non-test hardware parameters, such as the date and time, standby setting, screen contrast settings and printer setup. It is also used to access a directory of all stored tests, and the calculator function.

You can set up the other utility parameters, or change the utility settings, at any time except when an entry screen is displayed or when the instrument is carrying out a measurement.

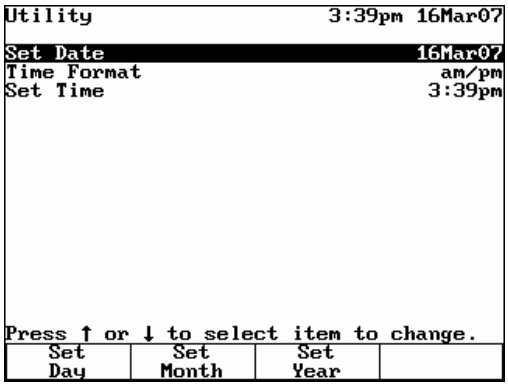
- Press *Utility*.



Setting the date and time

Follow these steps to access the date and time settings:

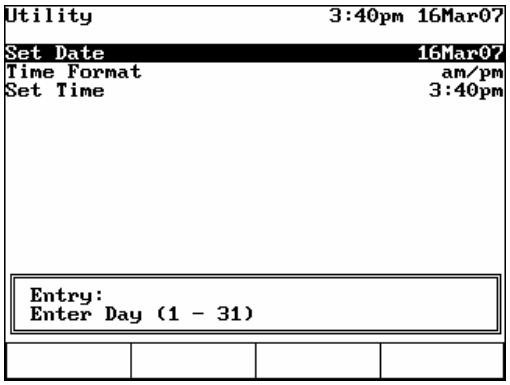
Highlight Date/Time Setup and press *Enter*.



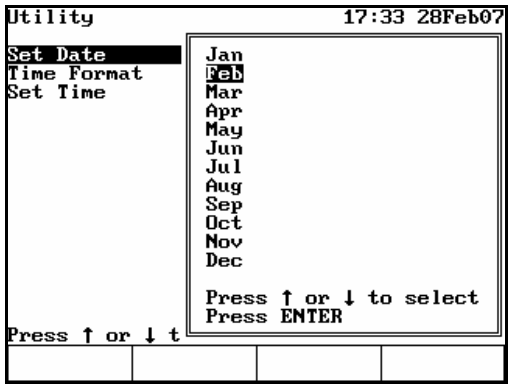
The screen displays the three date/time options that you can modify - date, time format and time.

To set the date:

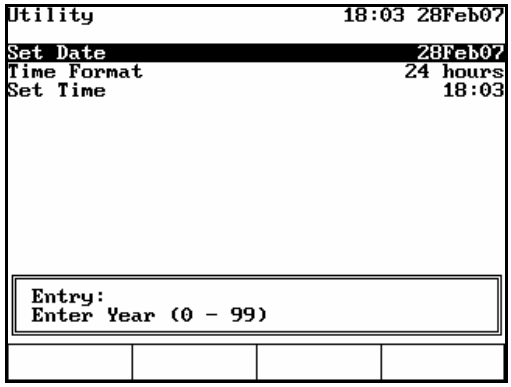
1. **Highlight Set Date and press *Enter*.**
2. **Press *Set Day*, type the date and then press *Enter*.**



3. Press *Set Month*, highlight the correct month and then press *Enter*.



4. Press *Set Year*, type the year and then press *Enter*.



5. When the date is correct, press *Esc* to save the settings and return to the Utility screen.

To select time format

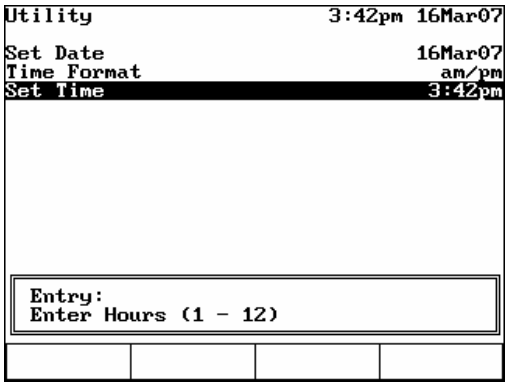
You can set up the spectrophotometer to display the time in either am/pm format or in 24-hour format.

To change the display format for the time, highlight Time Format and press *Enter* until the format that you want to use (AM/PM or 24 hour) appears.

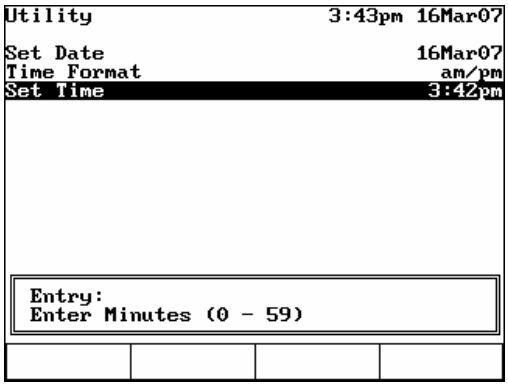
To set the time

1. **Highlight Set Time and press *Enter*.**

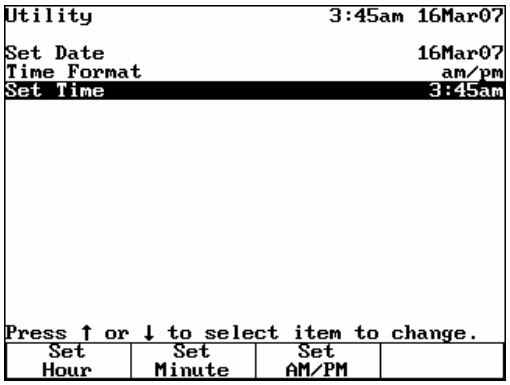
2. **To set the hour, press *Set Hour*, type in the hour and press *Enter*.**



3. **To set the minutes, press *Set Minute*, type in the minute and press *Enter*.**



4. **To select between AM and PM (if in AM/PM time format), press *Set AM/PM* until the appropriate setting appears.**

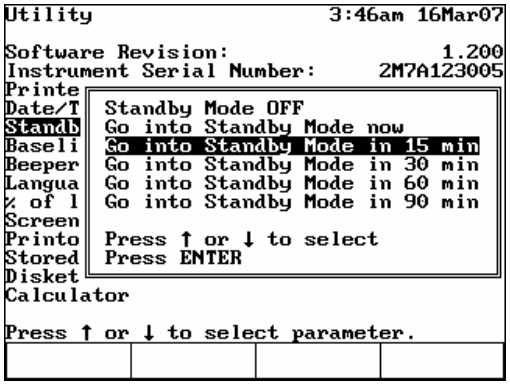


Note Any changes you make are saved automatically (even during power down) by battery backup. ▲

Selecting standby settings

To prolong xenon lamp life, your spectrophotometer was set at the factory to automatically go into standby mode after 15 minutes. To change Standby Mode time:

1. **Highlight Standby and press *Enter*.**


Utility 3:46am 16Mar07
Software Revision: 1.200
Instrument Serial Number: 2M7A123005
Standby Mode OFF
Go into Standby Mode now
Go into Standby Mode in 15 min
Go into Standby Mode in 30 min
Go into Standby Mode in 60 min
Go into Standby Mode in 90 min
Press ↑ or ↓ to select
Press ENTER
Calculator
Press ↑ or ↓ to select parameter.

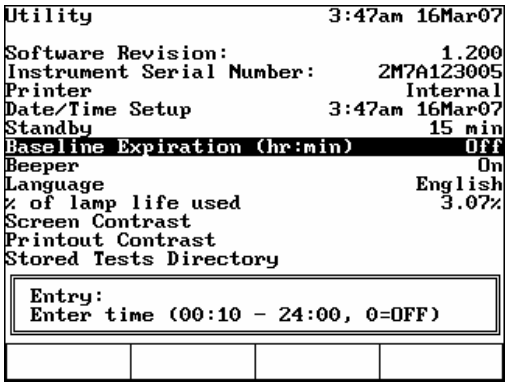
2. **Highlight the length of time you want the instrument will wait before entering standby mode and press *Enter*.**

Setting baseline expiration time

If you will be performing scans on your samples, you can set a time limit for which a collected baseline will be valid.

To set the baseline expiration time:

1. **Highlight Baseline Expiration (hr:min) and press *Enter*.**

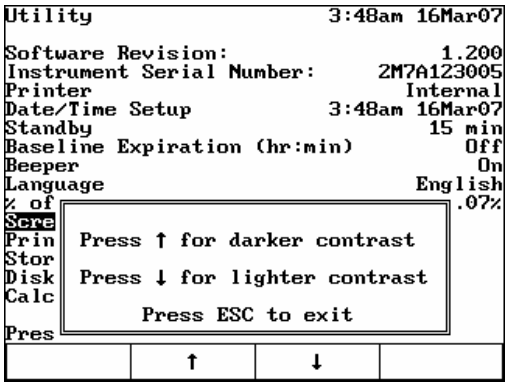


2. **Enter the desired time into the Entry baseline expiration time field and press *Enter*.**

Setting the screen contrast

To make it easier to read the display, you can adjust the screen contrast on the spectrophotometer.

1. **Highlight Screen Contrast and press *Enter*.**



2. **Adjust the screen contrast by following the instructions on the screen.**

3. **When the screen contrast is correct, press *Esc*.**

Setting up the internal printer

To set up the printer properly, you need to load the paper and set the utility parameters for the printer. Before setting up the parameters for the printer, be sure that the printer is installed. If you have ordered the internal printer as a separate item, you will need to install it.

1. **Install internal printer.**

   Refer to the "Installing the Internal Printer" section in the Accessories chapter for instructions on installing the printer.

2. **Load paper in the printer**

   Refer to the "Loading Paper in the Internal Printer" section in the Accessories chapter for instructions on installing the printer.

Setting the utility parameters for the printer

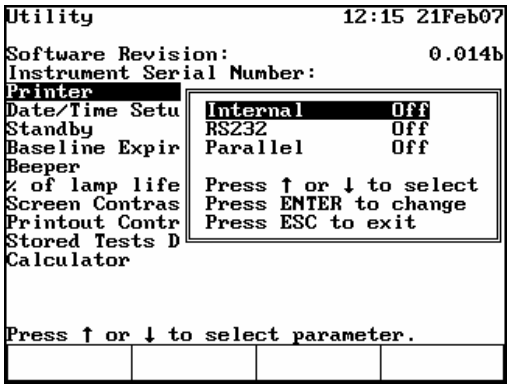
If you wish to use a printer you can output to:

- Internal printer
- External RS232 printer
- Parallel port to an HP PCL printer

To ensure that the spectrophotometer can output information correctly to the printer, you need to select the appropriate device.

1. **Press *Utility*.**

2. **Highlight Printer and press *Enter*.**



3. **Select the printer that you want to use and press *Enter* until On appears.**

4. **Press *Esc* to save the settings and return to the Utility screen.**

Note Text and graphics can be output via the internal printer and an external printer on the parallel port. Only text (no graphics) can be output via the RS232. ▲

Managing Stored Tests

The Evolution 60 spectrophotometer uses test files that contain the values for all the parameters needed to run a test, including the alignment of the cell changer and the other parameters for the accessories installed. Once you select the values for the parameters, you can assign a test name and save the test. You can then restore the test and run it without having to set up the parameters again.

When you power-down the instrument, the current test is maintained by battery back-up. This means that when you turn the instrument on again, the cell holder alignment and values for all parameters will be the same as they were when the instrument was last used. When you load a test that has been saved, the values for all parameters stored with that test will replace the current values for the test parameters again.

Software password

This password allows you to "lock" test setups (test parameters) so that they may not be overwritten or deleted. The password also allows you to remove the security so that you may edit the test parameters. Please refer to the Lock/Unlock section for more information on locking a test.

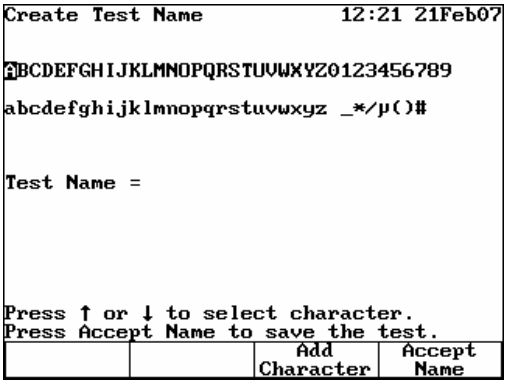
Note This password cannot be changed. ▲

Password: 4 3 6 3 7 9 7

Naming a test

When you save tests, you need to specify the name you want to use for the file. You can enter an alphanumeric name up to 16 characters long.

1. **After setting up the values for the test parameters highlight Test Name and then press *Enter*.**



You can use this screen to:

- Delete the name of a test
- Delete a character in the name of a test
- Add a character to the name of a test
- Accept the name of a test

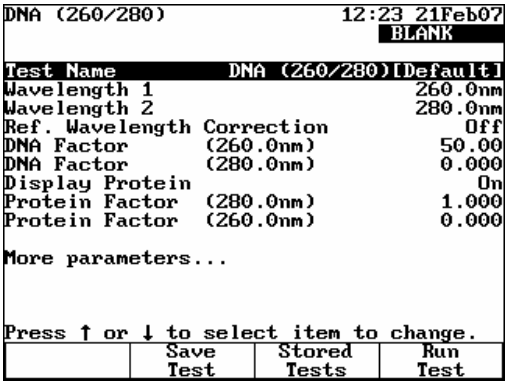
2. **Highlight the first character you want to use for the name of your test and press *Add Character* to add the selected character to the name.**

3. **Continue selecting and adding characters until you have selected all the characters for the name.**

4. **Press *Accept Name* to accept the name and return to the previous screen.**

Saving a test

Normally, you would save a test after you have entered the appropriate parameter values so you would be on a screen similar to the one shown below:



1. **Press *Save Test*.**

   The *Create Test Name* screen appears. If the desired test name is not displayed, enter it using the procedure in the Naming a Test section.

2. **If the desired name is displayed, press *Accept Name*.**

   A window allowing you to select whether the test is included as a SmartStart test (see the SmartStart section) is displayed.

3. **Highlight the appropriate test and press *Enter*.**

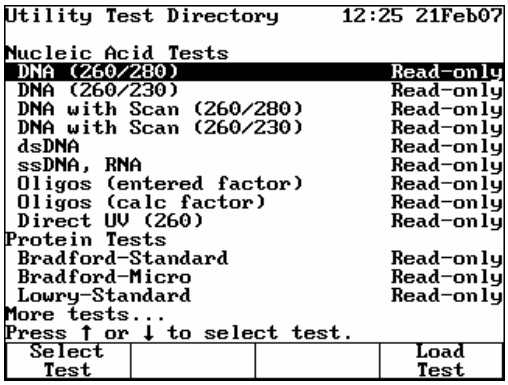
   The test is now saved in the instrument library.

Loading test files

Saved test files can be loaded from the internal memory from the Utility screen.

1. **To access all test files, press *Utility*.**

2. **Highlight Stored Test Directory and press *Enter*.**



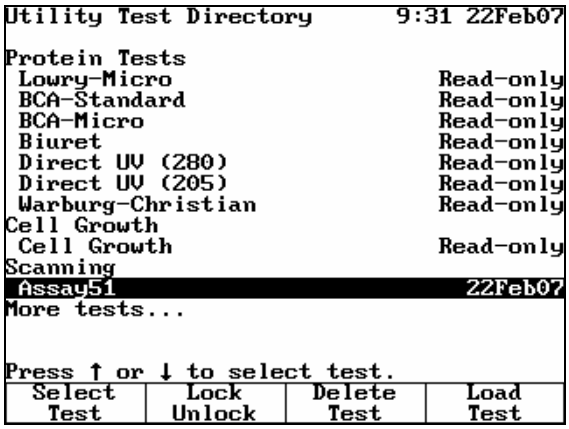
3. **Highlight the test you want to load and press *Load Test*.**

Note You can view the stored tests of a particular test type by pressing *Test*, selecting a test type, and pressing *Stored Tests*. ▲

Lock/Unlock

To lock or unlock a test, highlight the name of the test you want to lock or unlock and press *Lock/Unlock*.

Enter the password and press *Enter*.



Note If you want to lock or unlock access to the file, you must enter the software password of this manual. ▲

Delete Test

To delete a test, highlight the name of the test you want to delete and press *Delete Test*.

SmartStart

The SmartStart feature enables you to customize your spectrophotometer for your lab.

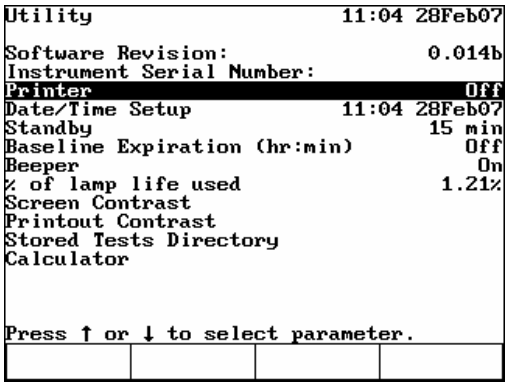
If you select one test as a SmartStart test, the instrument, when powered on, will automatically load this test and set up the instrument for immediate measurement.

If you select more than one test as SmartStart tests, the instrument, when powered on, will automatically display a menu that contains only those tests selected as SmartStart.

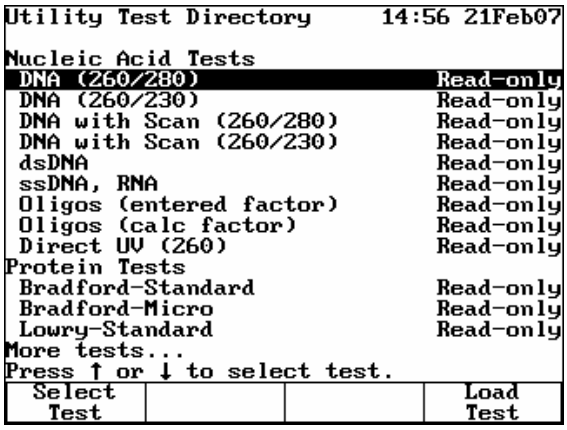
Note You will still be able to access the default main menu by pressing *Test*. ▲

Setting up a single test SmartStart

1. **Press *Utility* to display the Utility screen.**



2. **Highlight Stored Tests Directory and press *Enter*.**



3. **Highlight the appropriate test and press *Select Test* to add the test to the SmartStart menu.**

   An arrow sign ">" will indicate the test has been selected for the SmartStart menu.

   You can now either press *Esc* to return to the Utility screen, or you can power down the instrument.

To unselect a test:

- Press *Utility*.
- Highlight Stored Tests Directory and press *Enter*.
- Highlight test to be removed from SmartStart menu and press *Unselect Test* to remove the test from the SmartStart menu.
- The Main Menu will be displayed upon power up.

Setting up a multiple test SmartStart

1. **Follow steps 1 through 3 as outlined in Setting up a Single-test SmartStart.**

4. **Continue scrolling through the list and add tests until you have made all the appropriate selections.**

   An arrow sign ">" will indicate the tests selected for the SmartStart menu.

   You can now either press *Esc* to return to the Utility screen, or you can power down the instrument.

To remove tests from the SmartStart menu, see To Unselect a Test.

Concentration Units

Specifying concentration units

When you run concentration or kinetics tests, one of the test parameters is Units, which labels the results. The spectrophotometer includes a set of basic concentration units and you can also enter custom units if you wish.

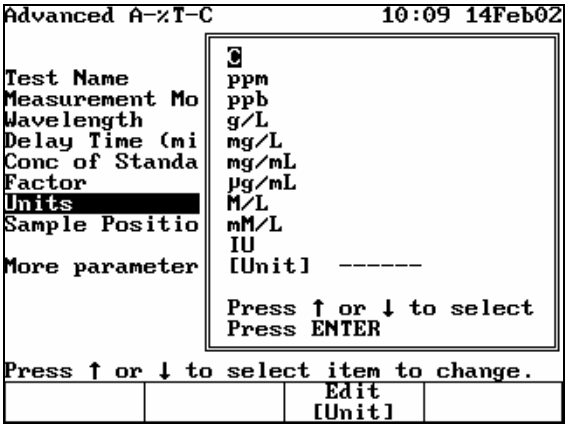
All programs in the spectrophotometer use the same list of basic units:

- C (concentration)
- ppm
- ppb
- g/L
- mg/L
- mg/mL
- ug/L
- M/L
- mM/L
- IU

In addition to these units, you can create your own custom unit, using a character list like the one described in Naming a Test. Once you create a custom unit, it will appear in the list that you use to select the units.

To select the units

1. **Highlight the Units parameter on the screen and press *Enter*.**



2. **Highlight the unit you want to select and press *Enter*.**

Creating custom units

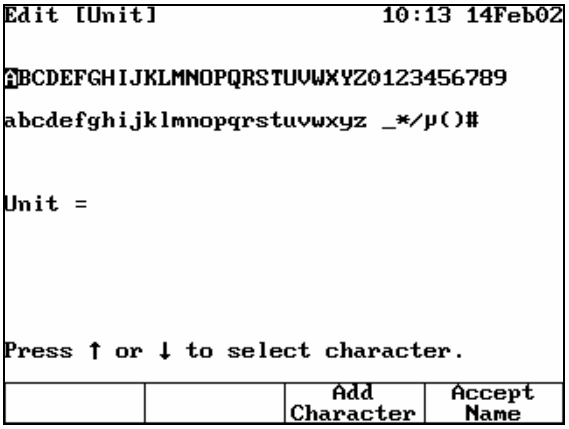
In addition to the basic concentration units, you can create a custom concentration unit and add it to the list. This custom concentration unit can be changed when desired.

Note Only one custom concentration unit is available in the list at one time. ▲

1. **With the Units Selection window displayed, press *Edit [Unit].***

   Use this screen to:

   - Delete the name of a unit
   - Delete a character in the name of a unit
   - Add a character to the name of a unit
   - Accept the name of a unit



2. **Follow steps 2 through 4 in the Naming a Test section above.**

   The name of the new custom unit appears on the list of basic units.

Cell Correction

Every test setup screen provides access to the cell correction program.

Note The Cell Correction program is not active from the Main (Basic ATC) screen. ▲

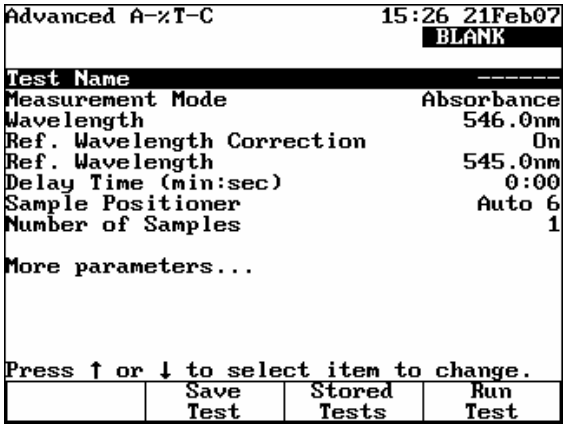
Note The Cell Correction feature is active only when the 6-Position Cell Holder is set to either Auto 6 or Auto 3. The feature is not active when the cell holder is set to 1-Cell or Manual 6, nor when the Single Cell Holder is installed. ▲

Before running the cell correction program:

- Clean the inside and outside of all the cells to be matched.
- Fill the cells with distilled water (or other blank solution), and place them in the sample compartment (see "Selecting and Positioning Cuvettes"). Be sure to place the blank cuvette in Cell "B" of the sample compartment.

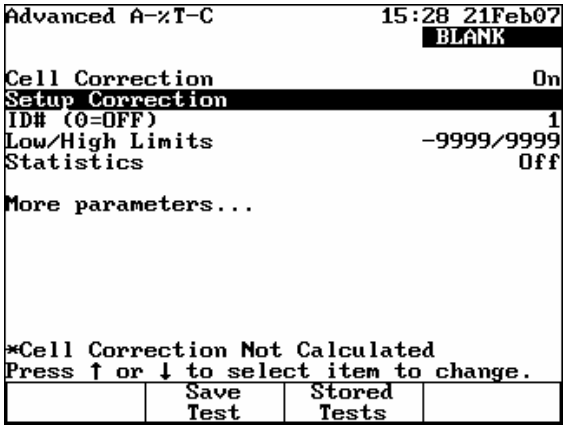
Running the Cell Correction program

1. **Load the test type or stored test.**



2. **If Cell Correction is not visible on the parameter list, then highlight *More parameters* and press *Enter*.**

3. **Highlight Cell Correction and press *Enter*.**



The Cell Correction function is now activated, as indicated by the word On across from Cell Correction on the test setup screen.

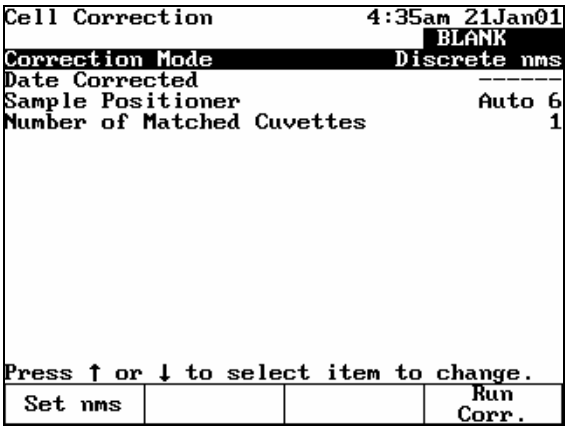
Note When Cell Correction is activated, additional parameter lines are added to the screen above the Cell Correction line. If the Cell Correction line is no longer visible on the screen, highlight More Parameters and press *Enter*. ▲

4. **Highlight Setup Correction and press *Enter*.**

5. **Highlight Correction Mode and press *Enter* to set the mode to either:**

   Scan – Cell Correction is run on a blank and one sample cell for the range of wavelengths you specify in Scanning mode.

   Discrete nms – The Cell Correction program is run on a blank and up to five sample cells for up to 31 user-specified, discrete wavelengths.



6. **If you selected Scan mode in the preceding step, specify the Start Wavelength and the Stop Wavelength values.**

7. **Press *Run Corr.* to start the Cell Correction program.**

   If you selected Discrete nms mode, first specify the wavelengths using the procedures which follow, and then the Cell Correction program.

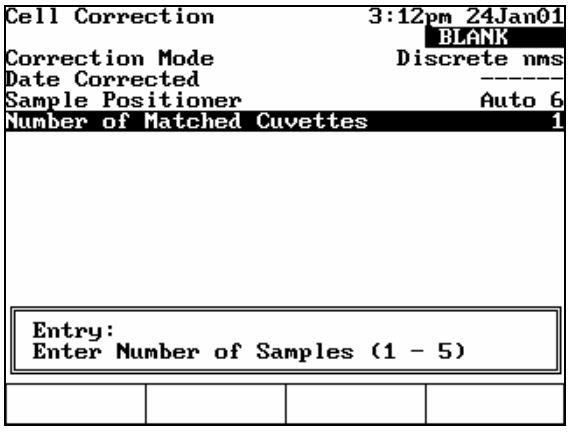
The Cell Correction program will measure the other cells against the blank and will record, store and date the measurements. From these measurements the Cell Correction program establishes the required correction factors, which then are automatically applied during all subsequent tests (if Cell Correction is activated).

Specifying wavelengths for Discrete nms mode

1. **Highlight Sample Positioner and press *Enter* to set this parameter to either Auto 3 (when using three large cell holders) or Auto 6 (when using six small cell holders).**

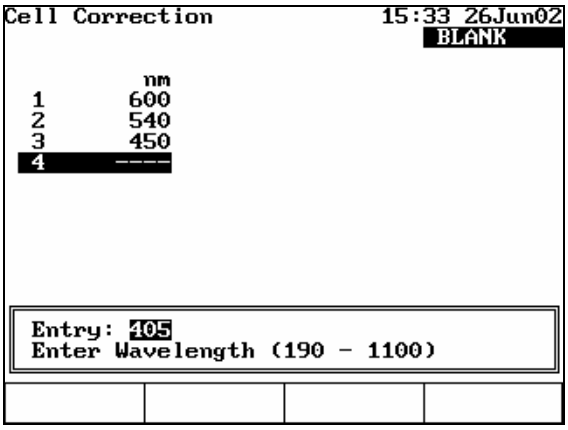
2. **Highlight Number of Matched Cuvettes and press *Enter*.**

3. **Specify the number of cells you are matching. Press *Enter*.**



4. **Press *Set nms* to select the wavelengths for which the Cell Correction program will be run.**

   A list of wavelengths appears.



Note Cells should be matched at all analytical wavelengths because matching at one wavelength does not guarantee matching at others. ▲

5. **Highlight the position where you want to enter the first wavelength.**

6. **Press *Add nm*.**

7. **Enter the value for the wavelength and press *Enter*.**

8. **Continue until you have entered all the wavelengths.**

After all the wavelengths have been entered, press *Run Corr.* to start the Cell Correction program. The Cell Correction program will measure the other cells against the blank and will record, store and date the measurements. From these measurements the Cell Correction program establishes the required correction factors, which then are automatically applied during all subsequent tests (if Cell Correction is activated).

Sample Positioner Setting

The spectrophotometer lets you use different cell holders and cell holder accessories to take measurements. When you select your test parameters, you will select the type of measurement you require and indicate how many samples you have. You can choose from the following measurement options:

Auto 6

Take one blank measurement and up to five sample measurements without changing the samples in the cell holder. The instrument **automatically** measures the blank (in the blank position), then **automatically** advances the cell holder to the appropriate position for the next measurement.

Auto 3

Take up to three measurements without changing the samples in the cell holder. The instrument **automatically** measures the blank (in the blank position), then **automatically** advances the cell holder to the appropriate position for the next measurement (position 2 or 4).

Single Cell Holder

Place the blank in the cell holder, measure it, place a sample in the cell holder, then measure your sample. This process is completely manual. In fact, the cell position buttons do not function when you select Single Cell Holder.

Note You can use the 6-Position Cell Holder as a single cell holder by selecting this option. However, measurement error can occur if, in the process of removing and inserting the cell, you move the position. Therefore, the Single Cell Holder is recommended for accessories requiring a high degree of positioning repeatability, such as the nanoCell accessory, small aperture microcells, the coupling module of fiber optics probes, and flowcells used with a sipper system. ▲

Manual 6

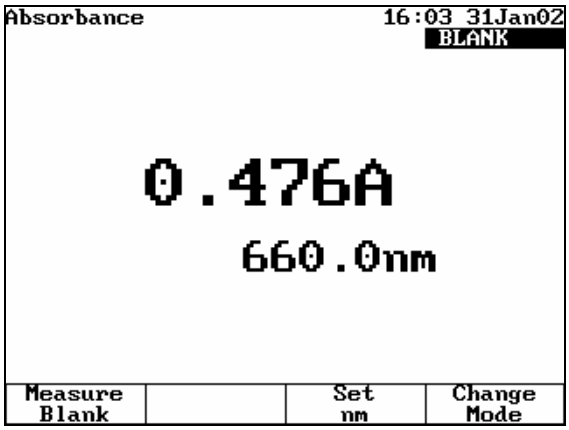
Take up to six measurements without changing the samples in the cell holder, using the cell position buttons to advance the cell holder to the appropriate position for the next measurement. You place the blank in the blank position and your samples in the other cell positions. Regardless of where the cell holder is positioned, when you press *Measure Blank* the cell holder automatically goes to the blank position and measures the blank. However, you can use the cell position buttons to select a different position for the measurement.

Note When you have the 6-Position Cell Holder installed, the instrument always considers the material in the B position as a blank. This means that even after measuring your blank the first time, you can place unknowns only in positions 1 through 5. ▲

Basic ATC Mode - Abs & %T Measurements

The Basic ATC mode puts the instrument into an 'instant measurement' mode. The lamp flashes continuously so that you can walk up to the instrument, insert your sample and measure it Depending on whether the mode is set to Absorbance (A), % Transmittance (%T), or Concentration, the result appears on the screen, along with the type of measurement, the date and time, the wavelength and the cell position used for the measurement.

To toggle between Absorbance, %Transmittance, and Concentration, press *Change Mode* until you see the desired mode is displayed. You can toggle from one mode to another whenever you see *Change Mode.*

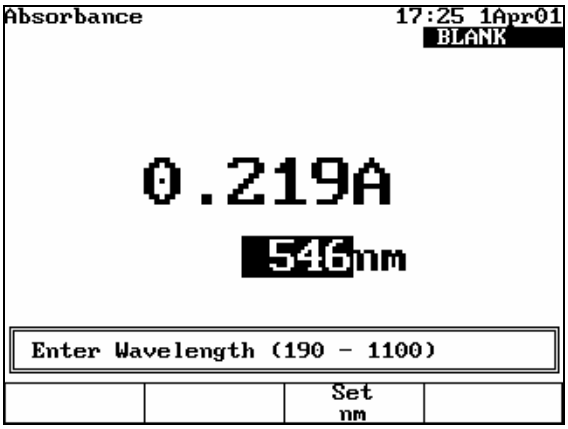


When Basic ATC is set to Absorbance or % Transmittance, you can perform these tasks:

- Set the wavelength
- Measure a blank
- Measure unknowns

Setting the wavelength

1. **Press *Set nm* or any number key to set the wavelength.**



2. **Enter the wavelength where you want the measurements taken, then press *Set nm* again.**

Measuring a blank

1. **Place the blank in the cell holder.**

   If your instrument has a 6-Position Cell Holder installed, be sure to place the blank in the B position.

2. **If you want to enter an absorbance or transmittance value for the blank, press a number key and enter the desired value in the Entry field.**

3. **Press *Measure Blank* to measure the blank.**

   When the instrument is finished measuring the blank, the message disappears.

Measuring unknowns

If you have the 6-Position Cell Holder installed, place the unknowns in the cell positions and press the corresponding cell position button to move the cell holder to the measuring position. The absorbance (ABS) or % transmittance (%T) measurement appears on the display.

If you have the Single Cell Holder installed, remove the blank and place the unknown in the cell holder. The absorbance or %Transmittance measurement appears on the display.

Basic Concentration measurements

Measuring concentration is similar to measuring Absorbance or %T. *Change Mode* can be used to switch to concentration measurements. The spectrophotometer allows you to measure concentration using either a factor or one standard to convert absorbance readings to concentration units.

- When you use a factor, you need to specify the factor and the concentration units.
- When you use a standard, you need to specify the concentration of the standard and measure its absorbance.

When Basic ATC is set to Conc/Std or Conc/Factor, you can perform these tasks.

- Set the wavelength
- Measure a blank
- Measure a standard or enter a factor
- Measure unknowns

The steps for taking measurements in the two modes are similar - the only difference will be whether you measure a standard or enter a factor.

Setting the wavelength and mode

1. **Press *Set nm* or any other number key to set the wavelength.**
2. **Enter the wavelength where you want the measurements taken and then press *Set nm* again.**
3. **Press *Change Mode* until the appropriate measurement mode (Concentration with Standard or Concentration with Factor) appears.**

Measuring a blank

1. **Place the blank in the cell holder.**

   If your instrument has a 6-Position Cell Holder installed, be sure to place the blank in the B position.

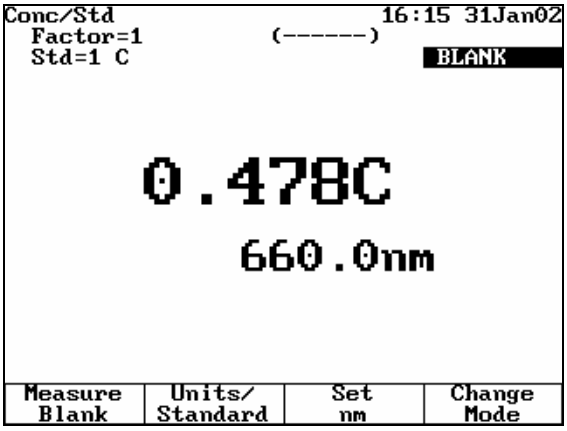
2. **Press *Measure Blank* to measure the blank.**

   When the instrument is finished measuring the absorbance of the blank, the message disappears.

Basic ATC Mode – Concentration Measurements

Using Conc/Std to measure concentration

1. **If necessary, press *Change Mode* to switch to the Concentration with Standard mode.**



   If your instrument has a 6-Position Cell Holder installed, be sure to place the blank in the B position, and the standard in position 1.

2. **Press *Measure Blank* to measure the blank.**

   If the Single Cell Holder is installed, remove the blank and place the standard in the cell holder.

3. **Press *Units/Standard* to set the units and measure the standard.**

4. **Press *Enter Conc*, then enter the concentration value of the standard and then press *Enter*.**

5. **Press *Select Units*, then highlight the appropriate unit in the list and press *Enter* to select the units for the concentration.**

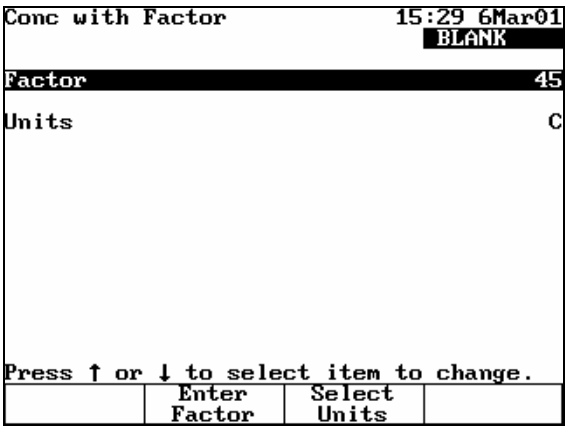
6. **Press *Measure Standard* to measure the standard.**

   When the instrument is finished measuring the absorbance of the standard, it displays the absorbance and calculated factor.

Using Conc/factor to measure concentration

1. **If necessary, press *Change Mode* to switch to the Conc With Factor mode.**

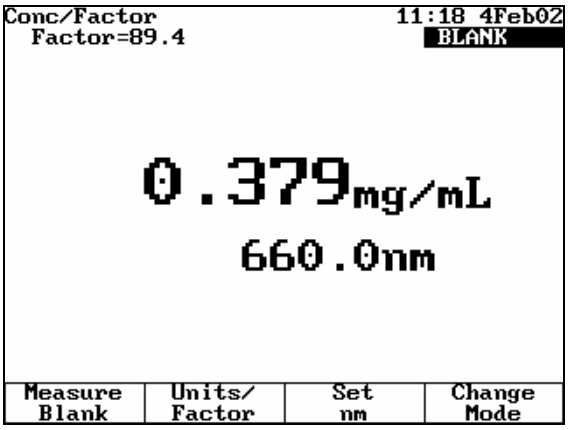
2. **Press *Units*/*Factor* to set the factor and select the units.**



3. **Press *Enter Factor*.**

4. **Type the desired factor value.**

5. **Press *Enter Factor* to accept the factor and return to the screen displaying the factor and units.**



6. **Press *Select Units*.**

7. **Highlight the appropriate unit in the list and press *Enter* to select the units for the concentration.**

8. **Press *Esc* to return to the Conc With Factor screen.**

Measuring unknowns

If the 6-Position Cell Holder is installed, place the unknown you want to measure in one of the cell positions and press the corresponding cell position button to move the cell holder to the measuring position.

The measurement appears on the display.

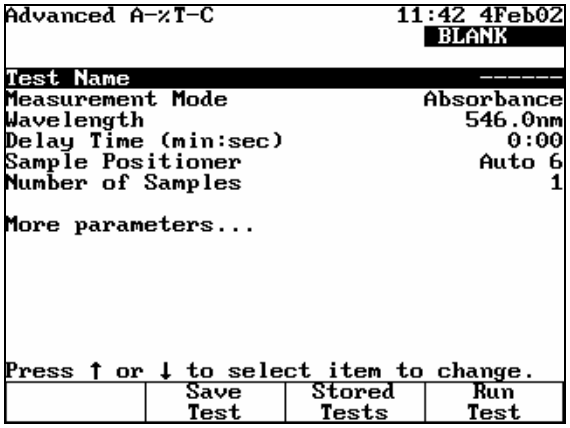
If the Single Cell Holder is installed, remove the blank and place the unknown in the cell holder. The measurement appears on the display.

Advanced A-%T-C— Abs & %T

When you use the Advanced A-%T-C program for Absorbance or %Transmittance measurements, you can perform these tasks:

- Select the measurement mode you want to use (Absorbance or %Transmittance)
- Run Cell Correction program
- Recall a test or set up new test parameters
- Measure a blank
- Measure unknowns

To get started, press *Test*. When the Test Types screen appears, highlight Advanced A-%T-C and press *Enter*.



Recalling a test

1. **From the Advanced A-%T-C screen, press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter*.**

   From this screen, you can:

   - Set up test parameters
   - Run Cell Correction program
   - Save a test
   - View the list of stored tests
   - Measure a blank and unknowns

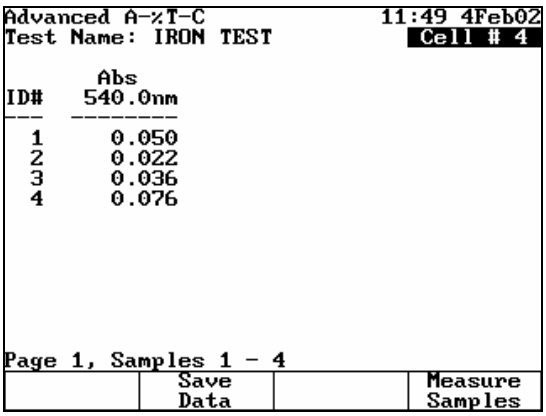
Setting up test parameters

1. **Highlight the name of the parameter you want to set.**

   Some parameters appear only if you select one of the concentration modes, while others appear regardless of the measurement mode you select. See the Parameters chapter for a complete list.

2. **When the parameters are set, press *Save Test* to save the test or *Measure Sample* to measure a blank or unknowns.**

Taking measurements



Taking measurements automatically (using Auto 6 or Auto 3)

1. **Press *Run Sample*.**

2. **Place the blank and the unknowns in the correct cell positions.**

3. **Press *Measure Sample* to measure the unknowns.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the sample measurements on the screen.

Taking measurements manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Sample*.**

   The Advanced A-%T-C measurement screen appears, prompting you to place your samples in the cell holder.

2. **Place the blank and unknowns in the cell holder.**

   If the 6-Position Cell Holder is installed, be sure to place the blank in the B position. You can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to measure the unknowns.**

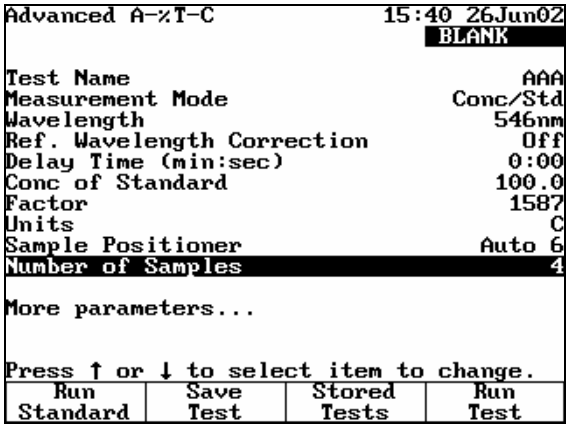
   The sample measurement appears on the screen. If a 6-Position Cell Holder is installed, press the cell position buttons to reposition the cell holder and measure the rest of the unknowns manually.

Advanced A-%T-C — Concentration

When you use the Advanced A-%T-C program for concentration measurements, you can perform these tasks:

- Select the measurement mode you want to use (concentration with one standard or concentration with a factor).
- Recall a test or set up new test parameters.
- Measure a standard or enter a factor (only if you select either concentration with one standard or concentration with a factor).
- Measure a blank and unknowns.

To get started, press *Test*. When the Test Types screen appears, highlight Advanced A-%T-C and press *Enter*.



Recalling a test

1. **Press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter* or *Load Test*.**

The parameters for the selected test appear on the screen.

Setting up test parameters

1. **Highlight the name of the parameter you want to set.**

   Some parameters appear only if you select one of the concentration modes, while others appear regardless of the measurement mode you select.

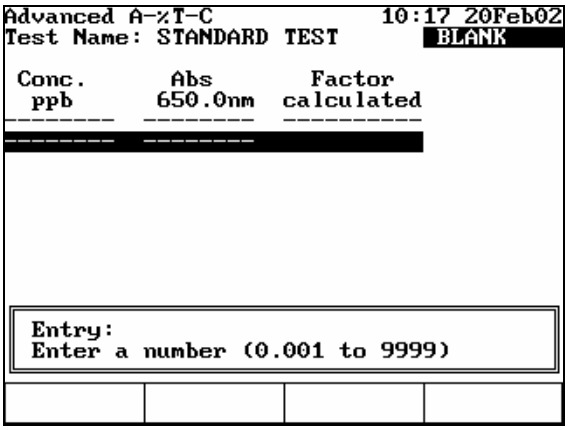
   See the Parameters chapter for a complete list.

2. **When the parameters are set, press *Save Test* to save the test or *Run Standard* to measure a standard (if in Conc/Std), or press *Run Test* (if in Conc/Factor).**

Measuring a standard

Measuring a standard automatically (using Auto 6 or Auto 3)

1. **With the Measurement Mode set to Conc/Std, press *Run Standard*.**



2. **Enter the concentration value of the standard and press *Enter*.**

3. **Press *Measure Blank*.**

4. **Place the blank and standard in the correct cell positions.**

5. **Press *Enter* to measure the blank and the standard.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the absorbance and calculated factor.

Measuring a standard manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Standard.***

2. **Enter the concentration value of the standard and press *Enter*.**

3. **Place the blank and standard in the correct cell positions.**

4. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

5. **Press *Measure Standard* to measure the standard.**

   When the instrument is finished measuring the absorbance of the standard, it displays the absorbance and calculated factor.

Entering a factor

Highlight Factor. If you need to change the factor, enter the correct factor.

If you need to change the units, highlight Units and select the correct units.

Measuring unknowns

Measuring unknowns automatically (using Auto 6 or Auto 8)

1. **Press *Run Test.***

   The Advanced A-%T-C measurement screen appears, prompting you to place your samples in the cell holder.

2. **Place the blank and unknowns in the correct cell positions and press *Enter*.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the sample measurements on the screen.

3. **Press *Measure Sample* to measure additional unknowns.**

Measuring unknowns manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Test*.**

2. **Place the blank and unknown in the correct cell position.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

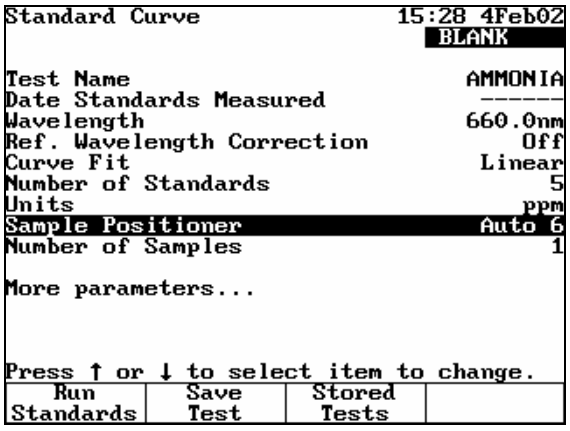
   If a 6-Position Cell Holder is installed, press the cell position buttons to reposition the cell holder and measure the rest of the unknowns manually.

Standard Curve

When you use the Standard Curve program, you can perform these tasks.

- Recall a standard curve or create a standard curve (set up the parameters and then measure the standards for the curve).
- Run the Cell Correction program.
- Measure unknowns.
- View the data - select between graphical and tabular displays.
- Edit a standard curve - change the number of standards, select a different curve fit or delete points from the curve.

  To get started, press *Test*. When the Test Types screen appears, highlight Standard Curve and press *Enter*.



Recalling a standard curve

1. **Press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter*.**

Setting the parameters for a standard curve

1. **Place the standards in the correct cell positions.**
2. **Set the parameters for measuring the standards.**

   See the Parameters chapter for a complete list.

   a. Enter the Test Name, Wavelength, Reference Wavelength Correction and Reference Wavelength.

   b. Select the Curve Fit, Units and Sample Positioner.

   c. Set the Number of Standards and Number of Samples.

   d. Enter the low and high limits.

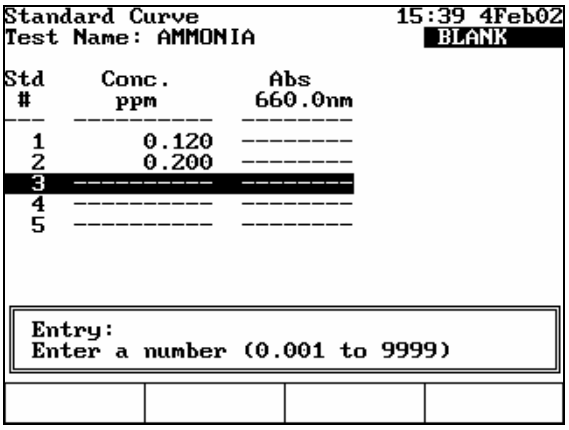
   e. Select the settings for the Statistics and AutoPrint functions.

   f. Run the Cell Correction program.

Measuring the standards for a standard curve

Measuring standards automatically (using Auto 6 or Auto 3)

1. **Place the blank and standards in the correct cell positions.**
2. **When all the parameters are correct, press *Run Standards* to set up and run the standards.**



3. **Enter the concentration value into the Entry concentration field and press *Enter*.**

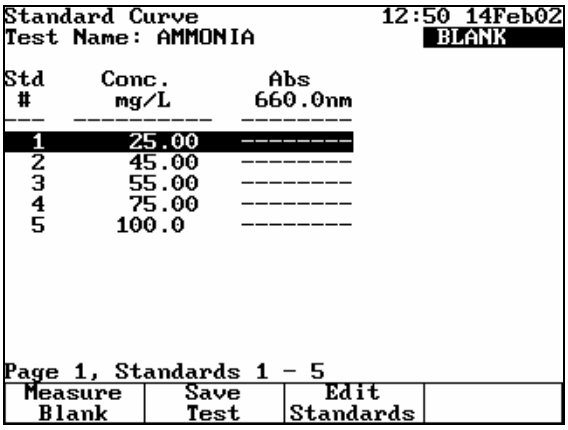
4. **Press *Measure Standards* to measure the blank and standards.**

   The instrument automatically measures the blank first and then measures the standards. When the instrument has measured all the standards, the Standards screen appears, showing the absorbance of each standard, along with the slope, intercept and correlation coefficient of the standard curve.

Measuring standards manually (using Manual 6 or Single Cell Holder)

1. **Place the blank and standards in the correct cell positions.**

2. **When all the parameters are correct, press *Run Standards* to set up and run the standards.**



3. **Enter the concentration value into the Entry concentration field.**

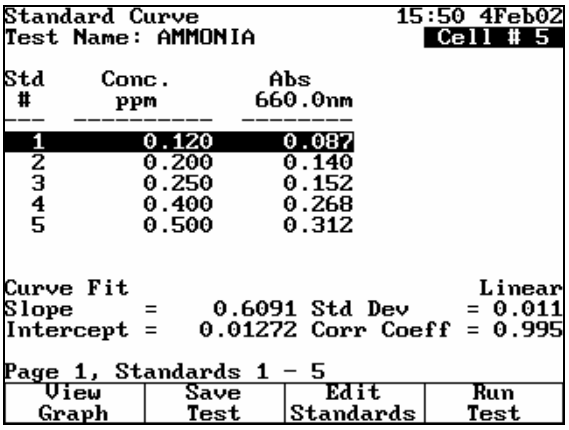
4. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

5. **Press *Measure Standards* to measure the standards.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the standards manually.

   When all of the standards have been measured, the Standards screen appears, showing the absorbance of each standard, along with the slope, intercept and correlation coefficient of the standard curve.



You can use this screen to:

- Display a graph of the standard curve data (press *View Graph*)
- Save the standard curve (press *Save Test*)
- Edit the standards (press *Edit Standards*)
- Measure your samples (press *Run Test*)

Measuring unknowns

Measuring unknowns automatically (using Auto 6 or Auto 3)

1. **Press *Run Test*.**

   The Samples screen appears.

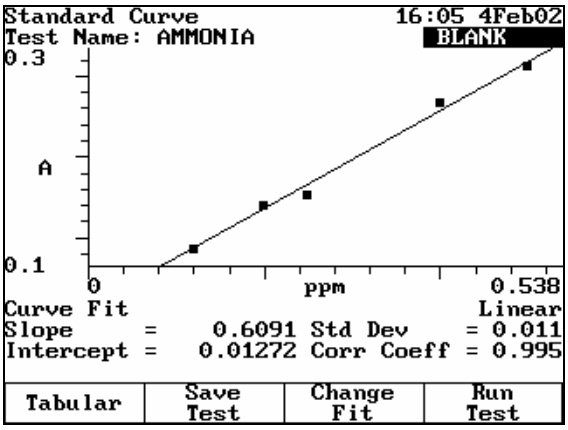
2. **Place the blank and unknowns in the correct cell positions.**

3. **Press *Enter* to measure.**

   The instrument automatically measures the blank first and then measures the unknowns.

   When the instrument has measured all the unknowns, the Standard Curve screen appears, showing the absorbance and concentration of each unknown.

   If you want to switch between tabular and graphical displays, press *View Graph/Tabular*.



Measuring unknowns manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Test.***

2. **Place the blank and unknowns in the correct cell positions.**

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank.

   When the blank has been measured, it returns to its previous cell position.

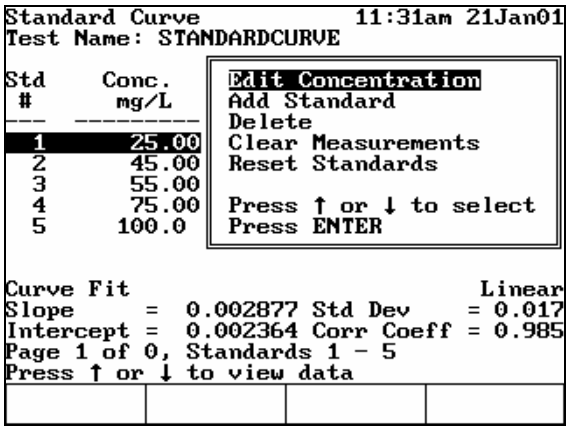
4. **Press *Measure Samples* to measure the unknowns.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

   When the instrument has measured all the unknowns, the Standards screen appears, showing the absorbance and concentration of each unknown.

Editing a standard curve

You may edit the concentration of any standard on a standard curve. In addition, you may change the number of standards, select a different curve fit or delete points from the curve.



To edit the concentration of a standard

1. **With the standard curve displayed on your screen, highlight the standard you want to edit and press *Edit Standards*.**
2. **With Edit Concentration highlighted, press *Enter*.**
3. **Press *Edit Conc* or a number key.**
4. **Enter the concentration value in the Entry field.**
5. **When the concentration value is correct, press *Enter* to accept the value.**

To add a standard

1. **With the standard curve displayed on your screen, press *Edit Standards*.**
2. **Highlight Add Standard.**
3. **Enter the concentration value of the additional standard in the Entry field.**
4. **When the concentration value is correct, press *Enter* to accept the value.**

5. **Press *Measure Standards* to remeasure all the standards.**

To delete a standard

1. **With the standard curve displayed on your screen, highlight the standard you want to delete and press *Edit Standards*.**
2. **Highlight Delete Standard and press *Enter* to delete the standard.**

To clear measurements

1. **With the standard curve displayed on your screen, press *Edit Standards*.**
2. **Highlight Clear Measurements and press *Enter*.**

   All the absorbance measurements will be removed from the screen.

To reset standards

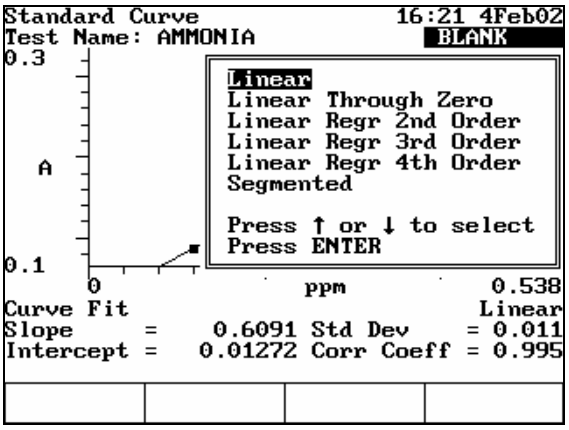
1. **With the standard curve displayed on your screen, press *Edit Standards*.**
2. **Highlight Reset Standards and press *Enter*.**

   All the standards and measurements will be removed.

To select a different curve fit for a standard curve

Note To change the curve fit for a standard curve, you must display the standard curve as a graph, not as a table. ▲

1. **With the standard curve you want to edit displayed as a graph on your screen, press *Change Fit*.**



2. **Highlight the curve fit you want to use for the standard curve and press *Enter*.**

   The instrument applies the selected curve fit to the data and displays the new fit on the graph.

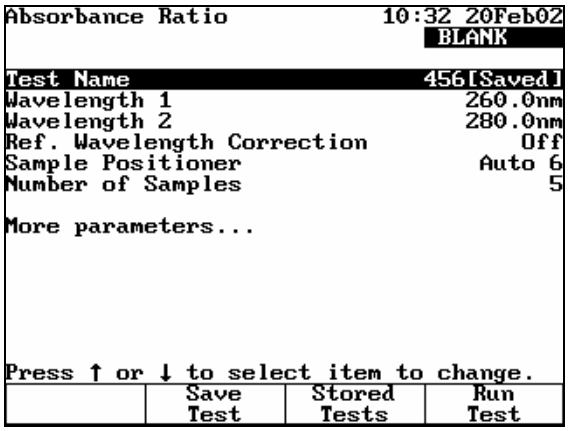
Absorbance Ratio

When you use the Absorbance Ratio program, you can perform these tasks:

- Recall a test *or* set up new test parameters
- Run the Cell Correction program
- Measure a blank
- Measure unknowns

To get started, press *Test*. When the Test Types screen appears, highlight Absorbance Ratio and press *Enter*.

Recalling a test



1. **With the Absorbance Ratio screen displayed, press *Stored Tests*.**

2. **Highlight the name of the test you want to recall and press *Enter*.**

From this screen, you can:

- Set up test parameters
- Run the Cell Correction program
- Save a test
- View the list of stored tests
- Measure a blank
- Measure unknowns

Setting up test parameters

1. **With the Absorbance Ratio screen displayed, highlight the name of the parameter you want to set.**

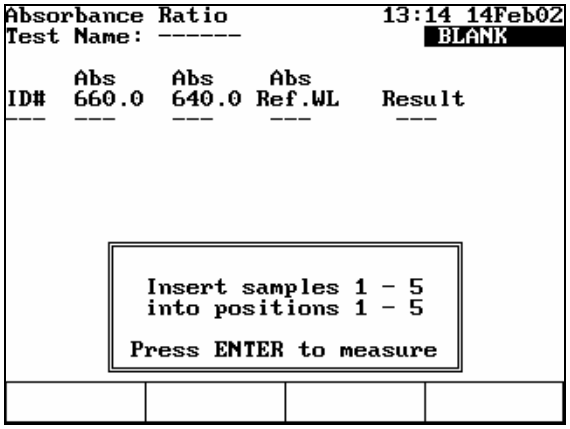
2. **When the parameters are set, press *Save Test* to save the test or *Run Test* to measure a blank or unknowns.**

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Note If Auto Save Data is ON, you must enter a Data File Name before you can access *Run Test* or *Measure Samples*. ▲

Measuring unknowns

Measuring unknowns automatically (using Auto 6 or Auto 3)



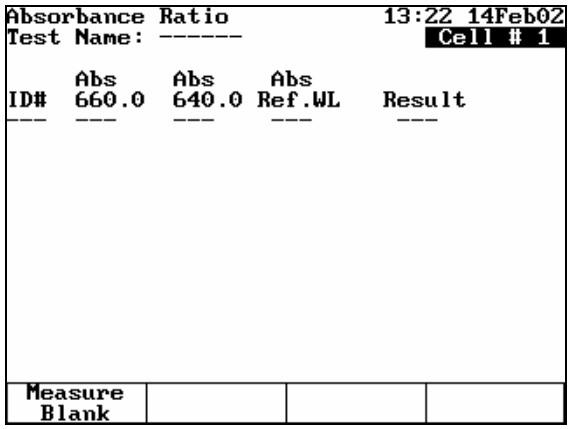
1. **Place the blank and unknowns in the correct cell positions.**

2. **Press *Enter* to start the measurement.**

   The instrument automatically measures the blank first, then measures the unknowns and then displays the sample measurements on the screen.

Measuring unknowns manually (using Manual 6 or Single Cell Holder)

1. **With the Absorbance Ratio screen displayed, press *Run Test*.**



2. **Place the blank and unknown in the correct cell position.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank.

   When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

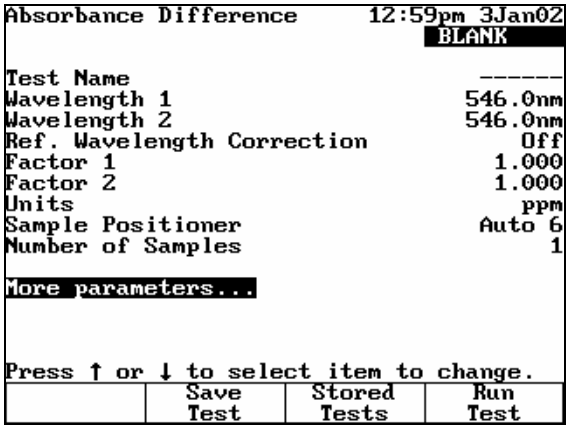
   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

Absorbance Difference

When you use the Absorbance Difference program, you can perform these tasks.

- Recall a test or set up new test parameters
- Run the Cell Correction program
- Measure a blank
- Measure unknowns

To get started, press *Test*. When the Test Types screen appears, highlight Absorbance Difference and press *Enter*.



Recalling a test

1. **With the Absorbance Difference screen displayed, press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter*.**

   From this screen you can:

   - Set up test parameters
   - Set up Cell Correction program

- Save a test
- View the list of stored tests
- Measure a blank
- Measure unknowns

Setting up test parameters

1. **With the Absorbance Difference screen displayed, highlight the name of the parameter you want to set.**

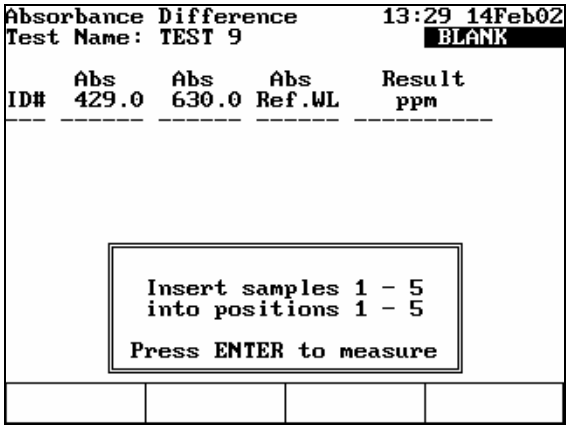
2. **When the parameters are set, press *Save Test* to save the test or *Run Test* to measure a blank or unknowns.**

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Note If Auto Save Data is ON, you must enter a Data File Name before you can access *Run Test* or *Measure Samples*. ▲

Measuring unknowns

Measuring unknowns automatically (using Auto 6 or Auto 3)


Absorbance Difference 13:29 14Feb02
Test Name: TEST 9
BLANK
ID# Abs 429.0 Abs 630.0 Abs Ref.WL Result ppm
Insert samples 1 - 5
into positions 1 - 5
Press ENTER to measure

1. **With the Absorbance Difference screen displayed, press *Run Test*.**

2. **Place the blank and unknowns in the correct cell positions.**

3. **Press *Enter* to start the measurement.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the sample measurements on the screen.

Measuring unknowns manually (using Manual 6 or Single Cell Holder)

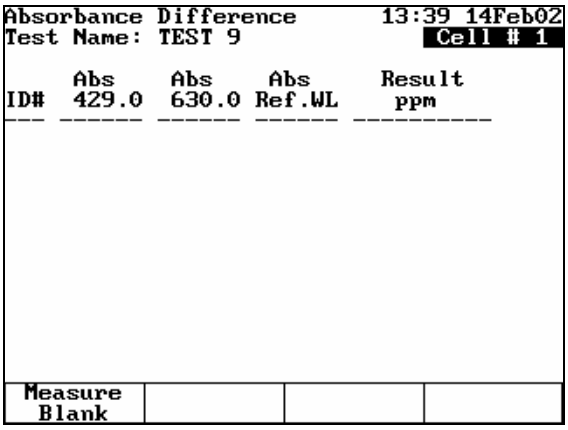
1. **With the Absorbance Difference screen displayed, press *Run Test*.**

2. **Place the blank and unknowns in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder. ▲

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position. ▲



4. **Press *Measure Sample* to start the measurement.**

   If a 6-Position Cell Holder is installed, press the position buttons to re-position the cell holder and measure the rest of the unknowns manually.

Kinetics

When you use the Kinetics program, you can perform these tasks:

- Recall a test or set up new test parameters
- Run the Cell Correction program
- Measure a blank
- Measure an unknown
- Recalculate reaction rates
- Modify the scale of the plot

When you use the Kinetics program, you can choose to work with either graphical data or with tabular data. You can perform the same functions regardless of the type of display you select. However, the location of the function keys varies depending on the type of display you choose.

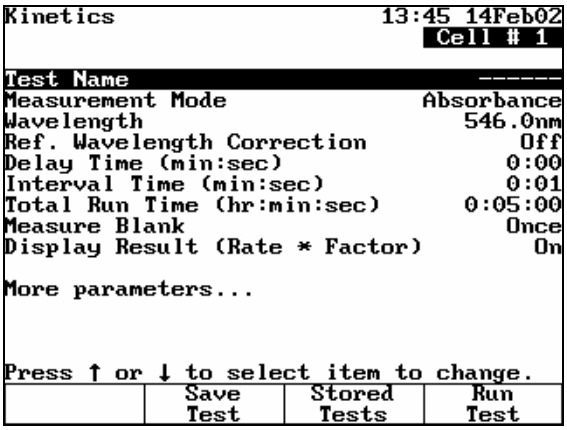
Note The Kinetics program allows you to measure only one unknown at a time. ▲

Note The Kinetics program allows you to collect up to 400 data points per run. When you set up your test parameters, be sure to select the interval time and total run time accordingly. ▲

To get started, press the *Test* button. When the Test Types screen appears, highlight Kinetics and press *Enter*.

Recalling a test

1. **With the Kinetics screen displayed, press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter***



From this screen you can:

- Set up test parameters
- Run the Cell Correction program
- Save a test
- View the list of stored tests
- Measure a blank
- Measure unknowns

Setting up test parameters

1. **With the Kinetics screen displayed, highlight the name of the parameter you want to set.**
2. **When parameters are set, press *Save Test* to save the test or *Run Test* to measure a blank or unknown.**

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Note If Auto Save Data is ON, you must enter a Data File Name before you can access *Run Test* or *Measure Samples*. ▲

Measuring unknowns

Note If a 6-Postion Cell Changer is installed, be sure to place the blank in position B and the unknown in cell position #1. The instrument always uses cell position #1 to scan the unknown. ▲

1. **With the Kinetics test parameters displayed on the screen, press *Run Test*.**

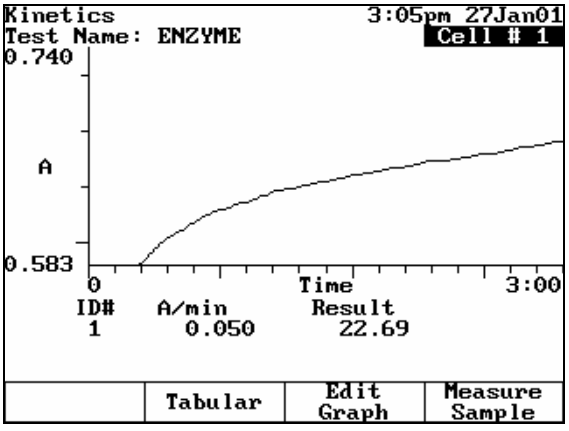
2. **If a 6-Position Cell Holder is installed, place the blank in position B and the unknown in position #1.**

3. **Press Measure Sample to measure the blank and the unknown.**

   When the instrument completes the measurement, the kinetics data and rate appear on the screen.

4. **If a Single Cell Holder is installed, press *Measure Blank* to measure the blank, then insert your unknown and press *Measure Sample*.**

   When the instrument completes the measurement, the kinetics data and rate appear on the screen.



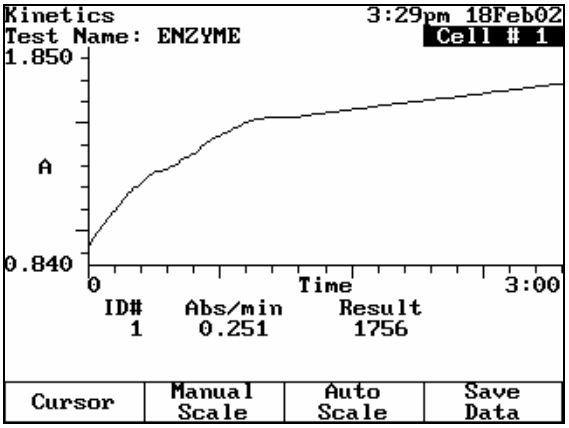
If you want to switch between tabular and graphical displays, press *Graph/Tabular*.

When you have a graphical display, you can press *Edit Graph* and then press *Cursor* to move the cursor line from one position to another on the plot. As the cursor moves the rate and result values indicate the values for the point on the plot where the cursor is located.

Recalling and recalculating graphical kinetics results

Within the Kinetics program, you can view and manipulate your results either in graphical form or in tabular form. When your results are displayed on the screen, you can modify the range (start and stop time) and the instrument automatically recalculates the reaction rate.

When you are working with graphical kinetics results, you need to press *Edit Graph* before you can rescale and recalculate.



You can modify the scale of your kinetics data plot in two ways - automatically or manually. When you select Auto Scale, the instrument automatically scales the X- and Y-axes so all the data appears on the plot. When you select Manual Scale, you select specific minimum and maximum values for the X- and Y-axes. Whenever you modify the scale, the instrument also automatically recalculates and displays the new reaction rate and result.

When the edit screen appears, you can:

- Use the Auto Scale function to change the scale, display the new graph and recalculate the results
- Use the Manual Scale function to change the scale, display the new graph and recalculate the results
- Use the cursor to select new minimum or maximum values for the X-axis and recalculate the results

Auto Scale function

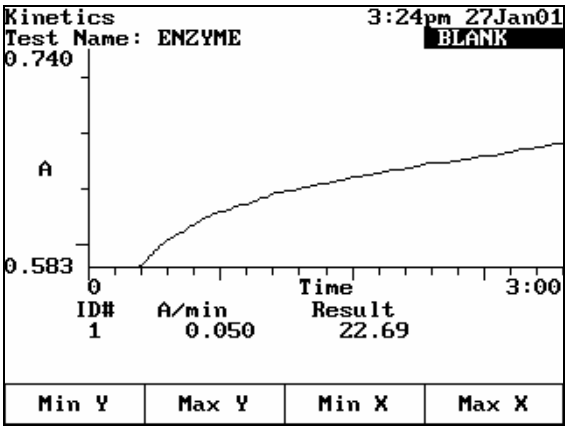
1. **With your kinetics data displayed on the Edit Graph screen, press *Auto Scale*.**

   The instrument automatically adjusts the minimum and maximum values for the X- and Y-axes so all the data appears on the plot. The instrument also recalculates the results, using all the data, and displays it on the screen.

Manual Scale function

1. **With your kinetics data displayed on the edit screen, press *Manual Scale*.**

   The manual scale options appear.



2. **Enter the appropriate minimum or maximum value for the X- or Y-axis and then press *Min Y, Max Y, Min X or Max X* to accept it.**

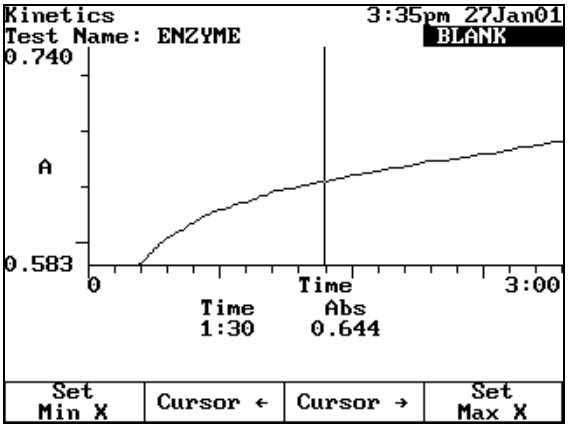
The instrument redraws the plot using the minimum and maximum value you have entered and displays the recalculated rate and result.

3. **Continue until you have entered all the values you want to change.**

Cursor function

1. **With your kinetics data displayed on the edit screen, press *Cursor*.**

   The cursor options appear.



2. **Press *Cursor* ← or *Cursor* → to position the cursor line on the appropriate point on the graph.**

   The instrument displays the data for the selected point.

3. **When the cursor line is in the correct position, press *Set Min X* or *Set Max X* to accept the selected point.**

   The instrument redraws the plot using the minimum and maximum value you have selected and displays the recalculated rate and result.

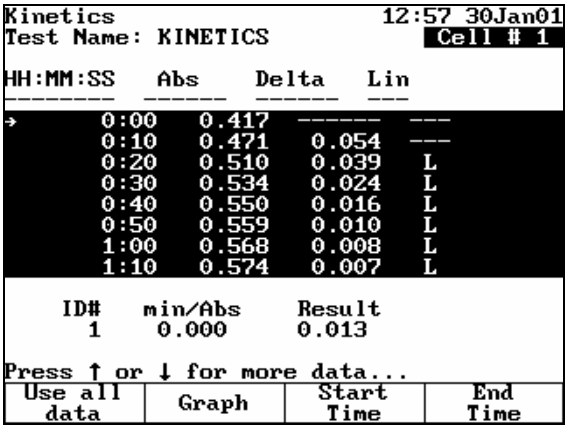
Rescaling and recalculating tabular kinetics results

When you are working with tabular kinetics results, you need to press *Edit Data* before you can rescale and recalculate.

After collecting your kinetics data, you can use all the data for the rate calculation or you can select specific start and end times. Whenever you modify the start and end times or select all the data, the instrument automatically recalculates and displays the new reaction rate and result.

When the edit screen appears, you can:

- Use all the data to recalculate the results
- Select specific start and end times for the rate calculation and recalculate the results



To use all the data to calculate the reaction rate

1. **With your table of kinetics data displayed on the edit screen, press *Use All Data*.**

   The instrument calculates the rate and displays it on the screen.

To select specific start and end times for the rate calculation

1. **With your table of kinetics data displayed on the edit screen, highlight the appropriate data point in the table.**

2. **Press *Start Time* or *End Time*.**

   The instrument displays the recalculated rate and result.

Scanning

When you use the Scanning program you can perform these tasks:

- Recall a test or set up new test parameters.
- Run the Cell Correction program
- Collect a baseline scan
- Scan an unknown
- View scan data
- Change the scale of the data plot
- Determine 3-point net measurements
- Calculate the area under a curve
- Label peaks and valleys

Note The Scanning program allows you to measure only one unknown at a time. Auto 6, Auto 3 and Manual 6 are not available for scanned measurements. ▲

Note If you want to set a baseline expiration time, press *Utility* and then highlight Baseline Expiration. Press *Enter* and set the desired time. ▲

To get started, press *Test*. When the Test Types screen appears, highlight Scanning and press *Enter*.

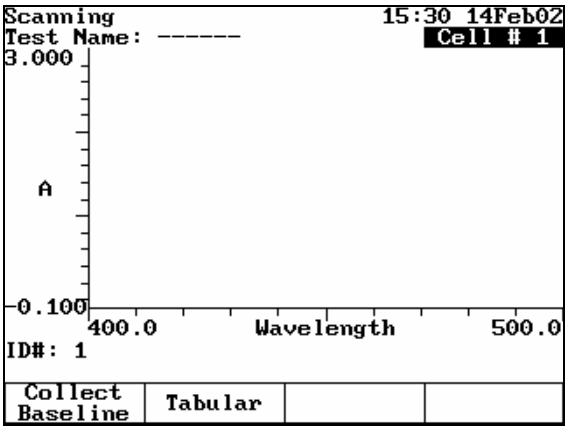
Recalling a test

1. **Press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter*.**

   The parameters for the selected test appear on the screen.

From this screen you can:

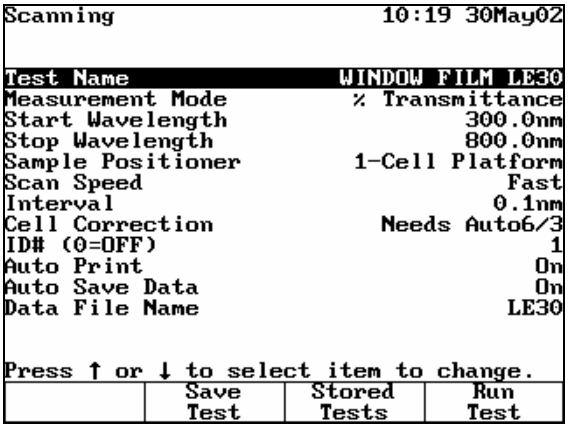
- Set up test parameters
- Set up Cell Correction program



Note If you want to switch between tabular and graphical displays, press *Graph*/*Tabular*. ▲

Setting up test parameters

1. **Highlight the name of the parameter you want to set.**
2. **When the parameters are set, press *Save Test* to save the test or *Run Test* to measure a blank or an unknown.**



Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

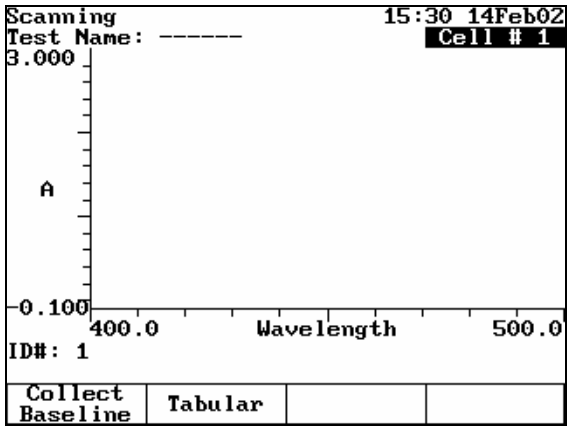
Note If Auto Save Data is ON, you must enter a Data File Name before you can access *Run Test* or *Measure Samples*. ▲

Collecting a baseline scan

Note If a 6-Position Cell Holder is installed, be sure to place the blank in the B position. The instrument always uses the B position to collect the baseline. ▲

1. **Press *Run Test*.**
2. **Place the blank in the B position.**
3. **Press *Collect Baseline* to collect the baseline.**

   When the instrument is finished measuring the blank, the message disappears.



Note To switch between tabular and graphical displays, press *Graph/Tabular*. ▲

Scanning an unknown

Note If a 6-Position Cell Holder is installed, be sure to place the unknown in cell position #1. The instrument always uses cell position #1 to scan the unknown. ▲

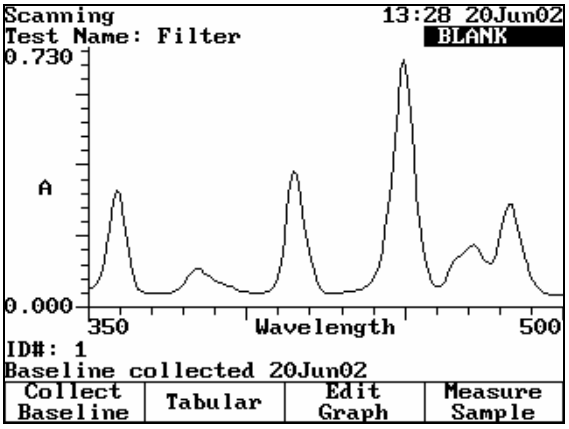
1. **Press *Run Test*.**

   If a 6-Position Cell Holder is installed, be sure to place the unknown in cell position #1.

2. **Press *Measure Sample* to measure the unknown.**

Note To switch between tabular and graphical displays, press *Graph/Tabular*. ▲

Note You may need to use the up and down arrow keys to view all the data for the scan. ▲



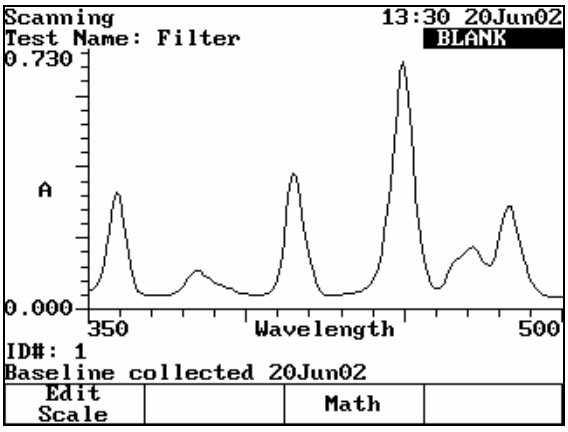
Viewing and manipulating scan data

Within the Scanning program, you can view and manipulate your results either in graphical form or in tabular form.

When you are working with graphical scan data, press *Edit Graph* before you perform other functions on the scan data.

When the edit graph screen appears you can:

- Rescale the graph
- Determine 3-point net values
- Calculate area under a curve
- Label peaks and valleys

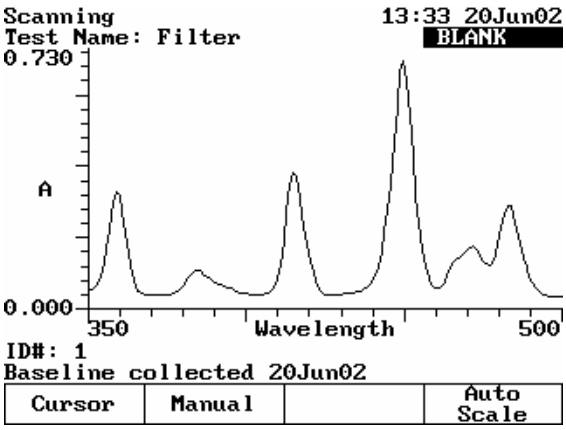


Rescaling graphical scan data

You can modify the scale of your scan data plot in two ways - automatically or manually. When you select Auto Scale, the instrument automatically scales the X- and Y-axes so all the data appears on the plot. When you select Manual Scale, you select specific minimum and maximum values for the X- and Y-axes. Whenever you modify the scale, the instrument also automatically recalculates and displays the new data plot.

Press *Edit Scale* to modify the scale. When the edit scale screen appears, you can:

- Use the *Auto Scale* function to change the scale and display the new graph.
- Use the *Manual Scale* function to change the scale and display the new graph.
- Use the cursor to identify specific points along the X-axis.

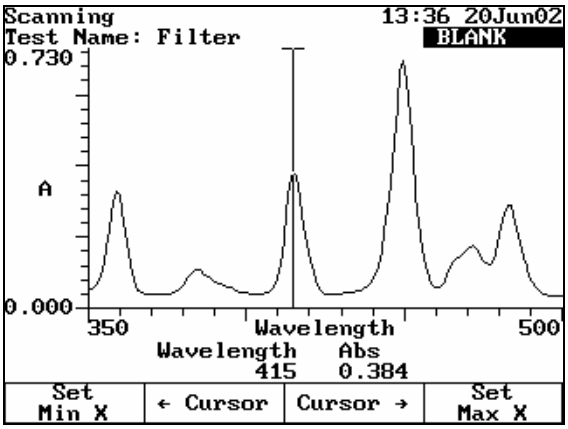


To use the Auto Scale function

With your scan data displayed on the edit scale screen, press *Auto Scale*. The instrument automatically adjusts the minimum and maximum values for the X- and Y-axes so all the data appears on the plot.

To use the Cursor

1. **With your scan data displayed on the edit scale screen, press *Cursor*.**



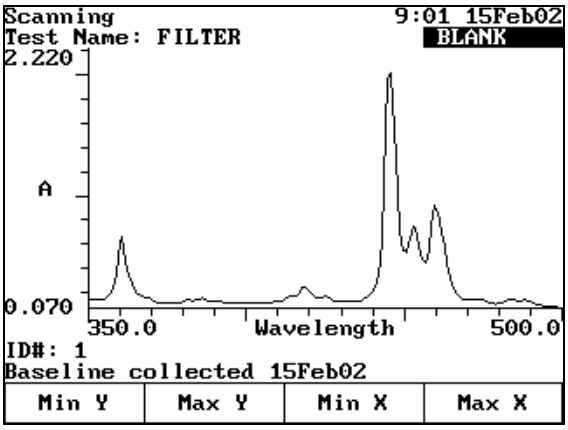
2. **Use *Cursor* ← to move to the desired minimum wavelength value. Press *Set Min X* to redraw the plot using the new minimum wavelength value.**

3. **Repeat using *Cursor* → and *Set Max X* to set the new maximum wavelength.**

To use the Manual Scale function

1. **Press *Manual Scale*.**

   The manual scale options appear.



2. **To set the minimum or maximum value for the X- or Y-axis, press *Min Y, Max Y, Min X or Max X*.**

3. **Enter the correct value and then press *Min Y, Max Y, Min X or Max X* to accept it.**

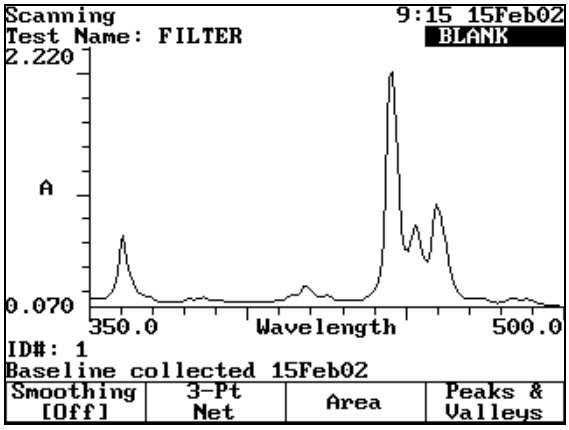
   The instrument redraws the plot using the minimum and maximum values you have entered.

Performing calculations on the scan data

You can modify your graph by performing calculations on the data. From the Edit Graph screen, press *Math*.

When the Math screen appears you can:

- Smooth data
- Determine 3-point net
- Calculate the area under a curve
- Label peaks and valleys



Smoothing data

If your scan shows sampling noise, you can smooth the data with the smoothing function.

1. **With your scan data displayed on the edit graph screen, press *Smoothing [On]*.**

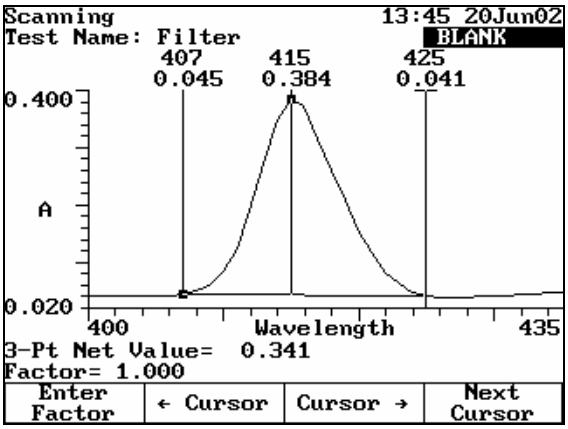
   The instrument performs a data smoothing function on the data and displays the smoothed graph.

Determining 3-point net measurements

1. **With your scan data displayed on the edit graph screen, press *Math*.**

2. **Press *3-Pt Net* to enter the 3-point net measurement screen.**

   A screen showing the cursor options and three cursor lines (designated for the left, center and right wavelengths) appears.



3. **Use *Cursor* → and *Cursor* ← to position the left cursor line to the desired wavelength value.**

   The instrument calculates the 3-point net absorbance for the selected wavelengths.

4. **Continue selecting the other wavelengths by pressing *Next Cursor* to activate the center and right cursor lines.**

   Select the wavelengths with *Cursor* ← and *Cursor* →.

   Repeat until all three wavelengths have been selected.

5. **Press *Enter Factor* to access the set factor box. Enter the desired factor and press *Enter*.**

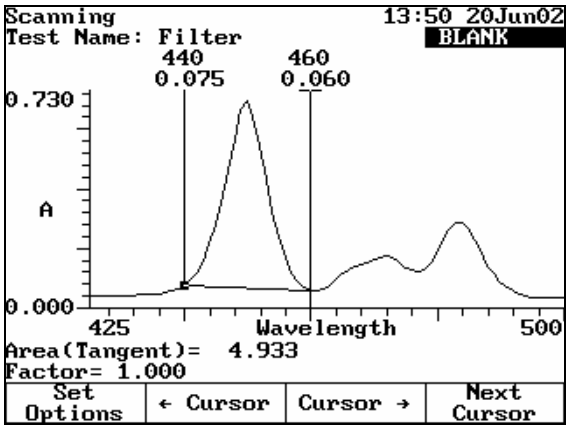
   The instrument calculates the value for the 3-point net absorbance for the selected wavelengths, multiplied by the selected factor.

Calculating the area under a curve

1. **With your scan data displayed on the edit graph screen, press *Math*.**

2. **Press *Area*.**

   The Area Under the Curve measurement screen appears.



3. **Use *Cursor* → and *Cursor* ← to position the left cursor line to the desired wavelength value.**

   The instrument calculates the area under the curve for the selected wavelengths.

4. **Continue selecting the other wavelengths by pressing *Next Cursor* to activate the next cursor line.**

   Select the wavelength with *Cursor* → and *Cursor* ←.

5. **Press *Set Options* to access the set options window.**

6. **Highlight *Factor*. Enter the desired factor and press *Enter*.**

7. **Highlight *Calculation* baseline.**

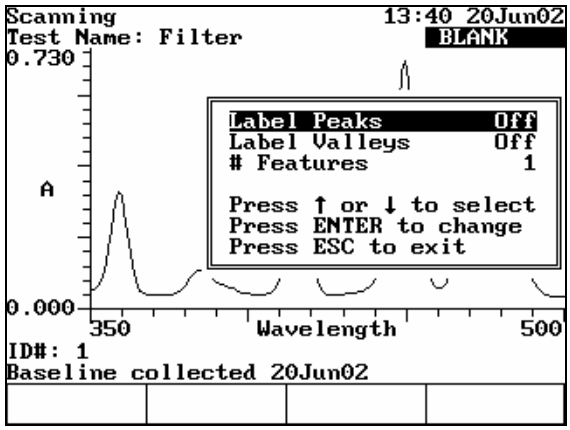
8. **Press *Enter* to toggle between Zero and Tangent.**

9. **Press *Esc* to return to the area under a curve screen.**

   The instrument calculates the area under a curve for the selected wavelengths, factor and calculation method.

Labeling peaks and valleys

1. **With your scan displayed on the edit graph screen, press *Math*.**



2. **Press *Peaks & Valleys*.**

   The Label Peaks and Valleys window appears.

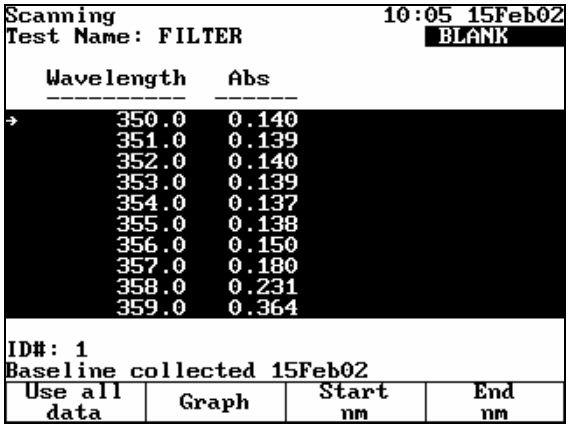
3. **Select the type of labels you want displayed and press *Enter*.**

   The instrument labels the selected items on your scan data plot.

Note Up to nine peaks or valleys can be calculated and displayed. ▲

Viewing and rescaling tabular scan data

When you are working with tabular scan data, you need to press *Edit Data* before you can perform other functions on the scan data.



To use all the scan data

With your table of scan data displayed on the edit screen, press *Use All Data*.

To select specific start and end wavelengths

1. **With your table of scan data displayed on the edit screen, highlight the appropriate data point in the table.**

2. **Press *Start nm* or *End nm*.**

   The instrument highlights the selected data points.

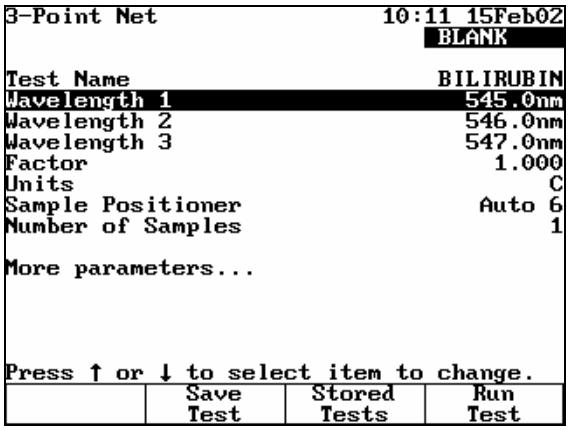
   You can press *Graph* to display the plot using the highlighted data points.

3-Point Net

When you use the 3-Point Net program, you can perform these tasks:

- Recall a test or set up new test parameters
- Run the Cell Correction program
- Measure a blank
- Measure unknowns

To get started, press *Test*. When the Test Types screen appears, highlight 3-Point Net and press *Enter*.



Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Note If Auto Save Data is ON, you must enter a Data File Name before you can access *Run Test* or *Measure Samples*. ▲

Recalling a test

1. **Press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter*.**

   The parameters for the selected test appear on the screen.

   From this screen you can:

   - Set up test parameters
   - Run the Cell Correction program
   - Save a test
   - View the list of stored tests
   - Measure a blank
   - Measure unknowns

Setting up test parameters

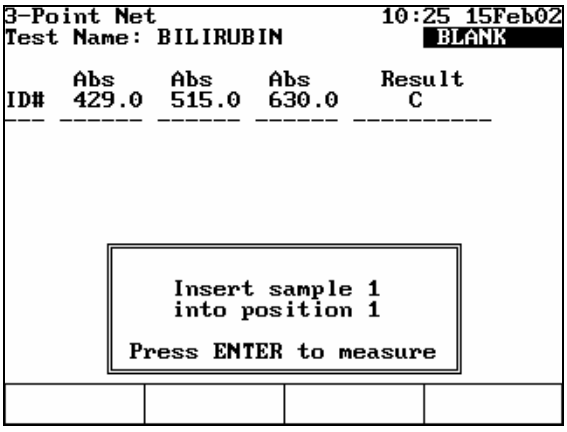
1. **Highlight the name of the parameter you want to set.**
2. **When the parameters are set, press *Save Test* to save the test or *Run Test* to measure a blank or unknowns.**

Taking measurements

Taking measurements automatically (Auto 6 or Auto 3)

1. **With the 3-Point Net Setup screen displayed and the parameters set, press *Run Test*.**

   The 3-Point Net measurement screen appears.



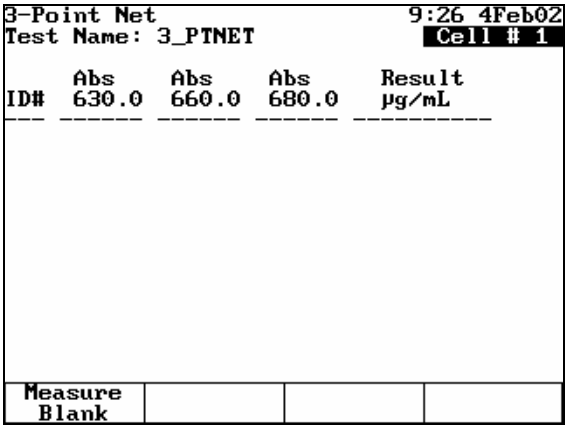
2. **Place the blank and the unknowns in the correct cell positions.**

3. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the sample measurements on the screen.

Taking measurements manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Test*.**



2. **Place the blank and unknowns in the cell holder.**

   If a 6-Position Cell Holder is installed, be sure to place the blank in the B position.

   You can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank.

   When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to measure the unknowns.**

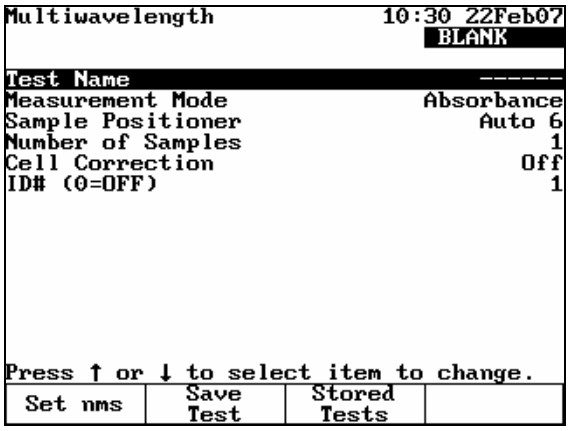
   If a 6-Position Cell Holder is installed, press the cell position buttons to reposition the cell holder and measure the rest of the unknowns manually.

Multiwavelength

When you use the Multiwavelength program, you can perform these tasks:

- Recall a test or set up new test parameters
- Run the Cell Correction program
- Add or delete wavelengths and factors
- Measure a blank
- Measure unknowns

To get started, press *Test*. When the Test Types screen appears, highlight Multiwavelength and press *Enter*.



Recalling a test

1. **Press *Stored Tests*.**
2. **Highlight the name of the test you want to recall and press *Enter*.**

   The parameters for the selected test appear on the screen.

   From this screen you can:

   - Set up test parameters

- Run the Cell Correction program
- Save a test
- View the list of stored tests
- Measure a blank
- Measure unknowns

Setting up test parameters

1. **With the Multiwavelength screen displayed, highlight the name of the parameter you want to set.**

   Refer to the procedures below for specific instructions on adding or deleting wavelengths and factors.

   If you have previously selected the wavelengths you wish to measure, press *Save Test* to save the test or *Run Test* to measure a blank or unknowns.

   If you have not selected the wavelengths, you can add wavelengths and factors as shown below.

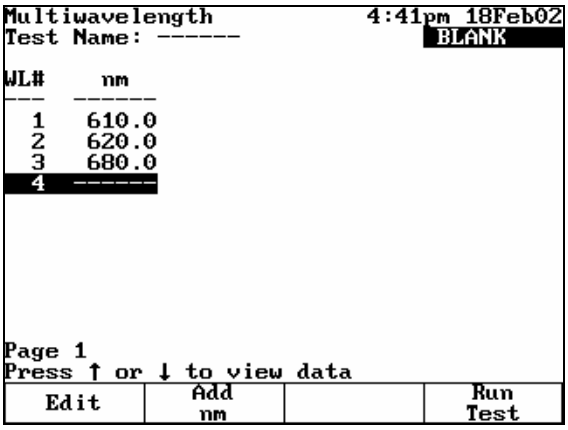
Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Note If Auto Save Data is ON, you must enter a Data File Name before you can access *Run Test* or *Measure Samples*. ▲

Adding wavelengths and factors

Note You can enter factors only when the measurement mode is set to Concentration/Factor. ▲

1. **Press *Set nms*.**



2. **Highlight the position where you want to enter the first wavelength and factor pair.**

3. **Press *Add nm*.**

4. **Enter the values for the wavelength and factor and press *Enter*.**

5. **When the values are correct, press *Add nm*.**

6. **Continue until you have entered all the wavelengths and factors.**

Deleting wavelengths and factors

1. **Highlight Set nms.**

Note If no wavelength values have been entered, the wavelength and factor columns will be empty. ▲

2. **Highlight the first wavelength and factor pair you want to delete.**

3. **Press *Delete nm*.**

Taking measurements

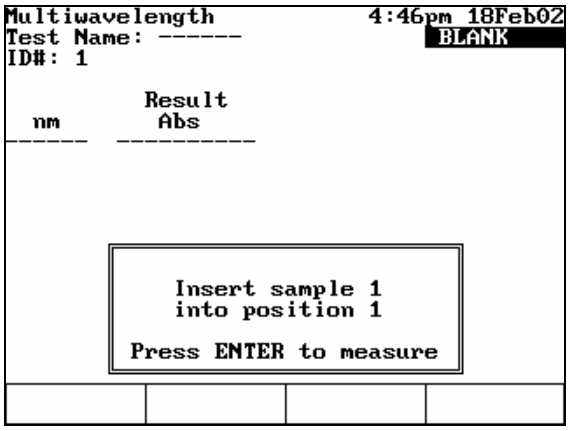
Taking measurements automatically (Auto 6 or Auto 3)

The Multiwavelength acquisition can be accessed from either the Set nms screen shown above or from the

Multiwavelength set up screen.

1. **Press *Run Test*.**

   The Multiwavelength measurement screen appears.



2. **Place the blank and the unknowns in the correct cell positions.**

3. **Press *Enter* to start the measurement.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the sample measurements on the screen.

Taking measurements manually (using Manual 6 or Single Cell Holder)

1. **With the Multiwavelength screen displayed and the parameters set, press *Run Test*.**

2. **Place the blank and unknowns in the cell holder.**

   If a 6-Position Cell Holder is installed, be sure to place the blank in the B position.

   You can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank.

When it completes the measurement, it returns to its previous cell position.

4. **With the list of wavelengths (and factors) displayed, press *Measure Samples* to measure the unknowns.**

   The instrument measures the absorbance at each wavelength and displays it on the screen.

   If you have set the measurement mode to Concentration/Factor, the instrument also displays the calculated concentration at each wavelength.

   If a 6-Position Cell Holder is installed, press *Cell Position* to reposition the cell holder and measure the rest of the unknowns manually.

Performance Verification

The Performance Verification program allows you to run tests to check the performance of your instrument.

Available tests include:

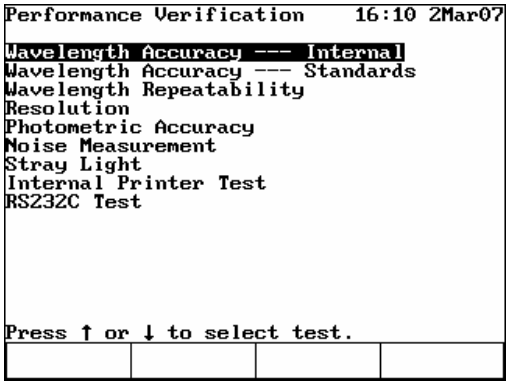
- Wavelength Accuracy – Internal
- Wavelength Accuracy – Standards
- Wavelength Repeatability
- Resolution
- Photometric Accuracy
- Noise Measurement
- Stray Light
- Internal Printer Test
- RS232C Test

Running the appropriate performance verification tests regularly and maintaining a log of your results helps document the reliability of the instrument and indicates potential performance issues.

Note If you have a printer installed and turned on, the instrument automatically prints out the test results for each performance verification test that you run. You can also press *Print* to print another copy of the results. ▲

Accessing the Performance Verification tests

1. **Press *Tests*.**
2. **Highlight Performance Verification and press *Enter*.**



Troubleshooting checklist

If a Performance Verification test fails, follow the instructions below to help you diagnose common problems that may cause a test to fail.

If a test continues to fail after you have tried all the recommendations in the list, follow the troubleshooting list for the test being run (included with the description of each test).

- Make sure you are following the instructions for the test properly.
- Make sure filters and standards are clean.
- Make sure the sample compartment door is closed while the test is running.
- Make sure the sample compartment is clear of any obstructions.
- Make sure the cell holder assembly is installed properly. If the 6-Position Cell Holder is installed, run the test once with the sample compartment door open to verify that the 6-Position Cell Holder is moving smoothly.

- Turn the instrument power OFF and then ON; verify that no problems are indicated by the power-on diagnostics.
- Make sure the lamp is ON.
- Make sure the lamp compartment is clear of any obstructions.

⚠ Warning Do not open the lamp compartment unless the instrument power is OFF. ▲

⚠ Warning Do not turn the instrument power ON unless the lamp compartment is closed. ▲

Wavelength Accuracy – internal

This test locates the peaks of the internal xenon lamp and displays the expected and measured wavelengths for the peaks.

- A xenon lamp has strong, fundamental lines at 229, 529 and 883 nm. These lines are an essential property of xenon and serve as a fundamental standard.

When running the internal standard test, remember that:

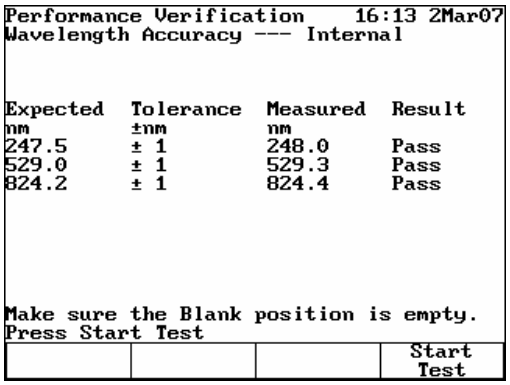
- The wavelengths and tolerance values are preset and cannot be changed.
- The cell holder should be empty.

1. **Highlight Wavelength Accuracy - Internal.**
2. **Press *Enter*.**

   The Wavelength Accuracy - Internal screen appears.

3. **Press *Start Test* to run the test.**

   The results of the test appear on the screen, indicating pass or fail for each wavelength.



If the test fails follow these guidelines:

- Repeat the test twice to verify that the test is failing consistently.
- Contact technical support for more troubleshooting advice.

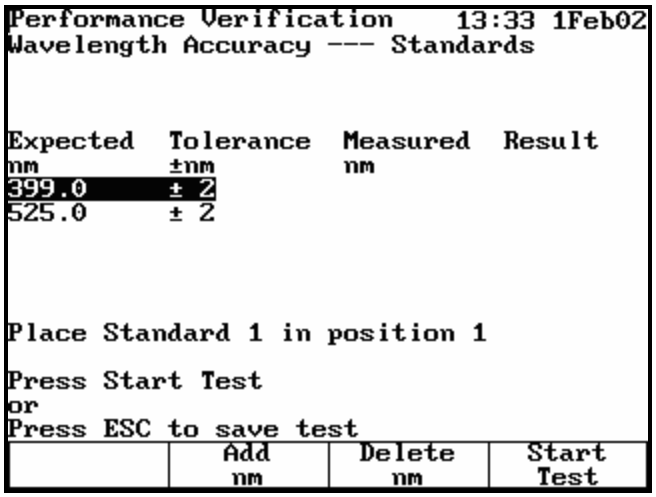
Wavelength Accuracy - Standards

This test measures the absorbance or %T of a wavelength accuracy standard at up to five wavelengths and compares the results with specified tolerances. The wavelengths and tolerances are preset, but you should change them to the values on the calibration certificate included with your standards.

When running the Wavelength Accuracy test, remember that:

- You will need Wavelength Accuracy standards that are designed to measure the wavelength accuracy at the specified wavelengths.
- An empty cell holder should be used as the blank.
- The standards need to be measured in the order that they are listed on the test screen.
- 1-5 standards can be used.

1. **Highlight Wavelength Accuracy - Standards and press *Enter*.**

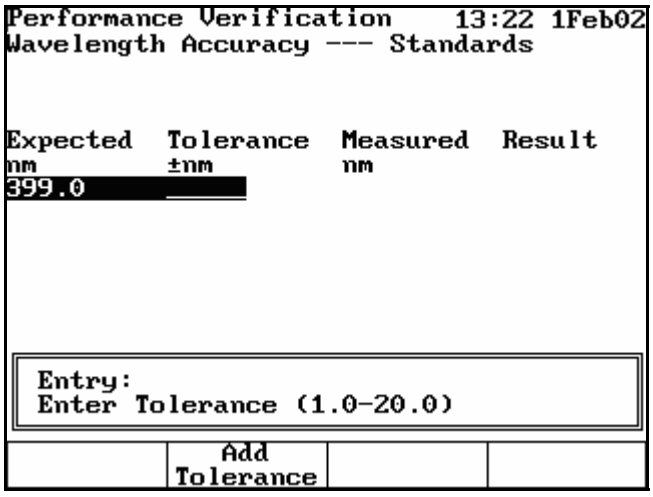


Adding wavelengths

1. **If you need to add a wavelength, press *Add nm* and enter the wavelength value in the Entry field.**

2. **When the value is correct, press *Add nm* again to add the wavelength to the list.**

   The screen changes to prompt you to enter the tolerance for the wavelength you just entered.



3. **Enter the tolerance value in the Entry field.**

4. **When the value is correct, press *Enter* to add the tolerance value to the list.**

Deleting wavelengths

1. **If you want to delete a wavelength, highlight the appropriate wavelength.**

2. **When the appropriate wavelength is highlighted, press *Delete nm* to delete the wavelength.**

Running the test

1. **Verify that the wavelengths and tolerances are set correctly.**

2. **Press *Start Test* to run the test.**

   The results of the test appear on the screen, indicating pass or fail for each wavelength.

If the test fails follow these guidelines:

- Repeat the test twice to verify that the test is failing consistently.
- Make sure the target value you entered for the calibrated wavelength is the same as the value printed on the calibration certificate for the standard.
- Make sure the tolerance value you entered for the calibrated wavelength is the same as the value given on the calibration certificate for the standard.

Wavelength Repeatability

This test measures the ability of the spectrophotometer to return to an identical wavelength in a repeatable manner. The test uses the internal xenon lamp.

- A xenon lamp has a strong, fundamental line 529 nm. This line is an essential property of xenon and serves as a fundamental standard.

When running the internal standard test, remember that:

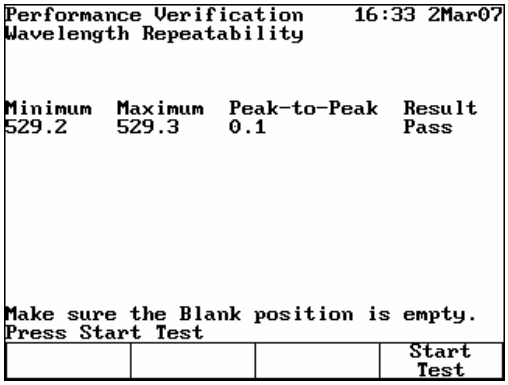
- The wavelength and tolerance values are preset and cannot be changed.
- The cell holder should be empty.

1. **Highlight Wavelength Repeatability.**
2. **Press *Enter*.**

   The Wavelength Repeatability screen appears.

3. **Press *Start Test* to run the test.**

   The results of the test appear on the screen, indicating pass or fail for each wavelength.



If the test fails follow these guidelines:

- Repeat the test twice to verify that the test is failing consistently.
- Contact technical support for more troubleshooting advice.

Resolution

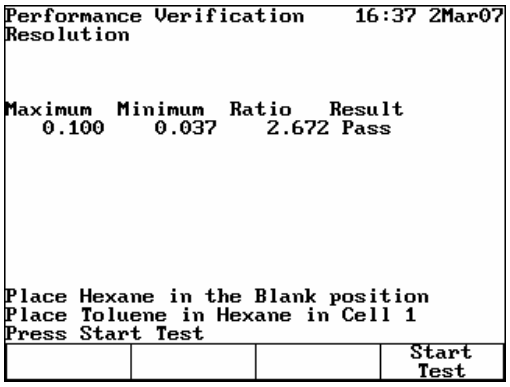
This test measures the ability of the spectrophotometer to resolve adjacent features in a spectrum. The test is performed using a 0.02% (v/v) solution of toluene in hexane and requires a hexane blank.

When running the internal standard test, remember that the wavelengths and tolerance values are preset and cannot be changed.

1. **Highlight Resolution.**

2. **Press *Enter*.**

   Ensure that Hexane is in the blank position and Toluene in Hexane is placed in Cell 1.



3. **Press *Start Test* to run the test.**

   The results of the test appear on the screen, indicating pass or fail for each wavelength.

If the test fails, follow these guidelines:

- Repeat the test twice to verify that the test is failing consistently.
- Contact technical support for more troubleshooting advice.

Photometric Accuracy

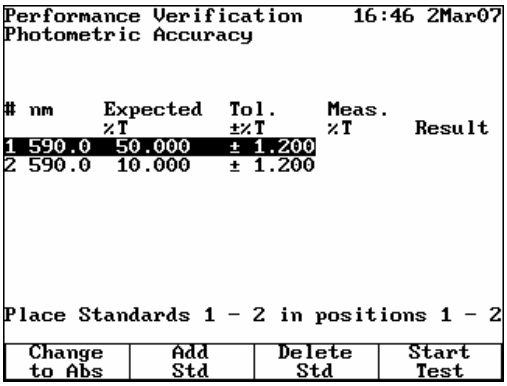
This test measures the absorbance (or %T) of a set of standards and compares the results with specified tolerances. The wavelength absorbencies and tolerances are preset, but you should change them to the values on the calibration certificate included with your standards.

Note You can display the tolerances for this test in either absorbance or %Transmittance. ▲

When running the Photometric Accuracy test, remember that:

- You will need Wavelength Accuracy standards that are designed to measure the wavelength accuracy at the specified wavelengths.
- An empty cell holder should be used as the blank.
- The standards need to be measured in the order that they are listed on the test screen.
- 1-5 standards can be used.

1. **Highlight Photometric Accuracy.**
2. **Press *Enter*.**


Performance Verification 16:46 2Mar07
Photometric Accuracy
nm Expected %T Tol. ±%T Meas. %T Result
1 590.0 50.000 ± 1.200
2 590.0 10.000 ± 1.200
Place Standards 1 - 2 in positions 1 - 2
Change to Abs
Add Std
Delete Std
Start Test

Selecting the mode

To switch between absorbance and %Transmittance, press *Change to %T* (or *Change to ABS*) until the appropriate mode appears.

Adding standards

You will need to set three values whenever you add a standard - the wavelength, the absorbance (or %Transmittance) and the tolerance value.

1. **If you need to add a standard, press *Add Std* and enter the wavelength value in the Entry field.**

2. **When the wavelength value entry is complete, press *Enter* or *Add nm* to add the wavelength to the list.**

   The screen changes to prompt you to enter the absorbance (or %T) for the wavelength you just entered.

3. **Enter the %T value in the Entry field.**

4. **When the %T value entry is complete, press *Enter* to add the %T value to the list.**

   The screen changes to prompt you to enter the tolerance value for the wavelength you just entered.

5. **Enter the tolerance value in the Entry field.**

6. **When the tolerance value entry is complete, press *Enter* to add the tolerance value to the list.**

   The screen returns to the test screen displaying the values you just entered for that standard.

7. **Press *Start Test* to begin the measurement or Press *Esc* to save test.**

Deleting standards

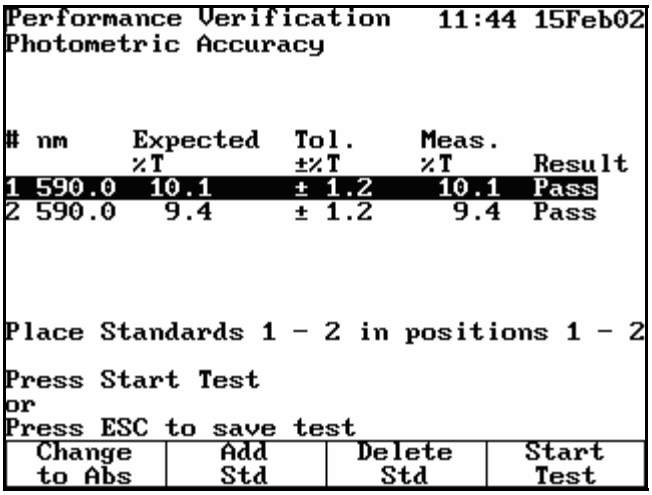
1. **If you need to delete a standard, highlight the appropriate standard.**
2. **Press *Delete Std* to delete the standard from the list.**

Running the test

With the Photometric Accuracy screen displayed, make sure that the wavelengths, absorbance (or %T) values and tolerances are set correctly.

1. **Press *Start Test* to run the test.**

   The results of the test appear on the screen, indicating pass or fail for each wavelength.


Performance Verification 11:44 15Feb02
Photometric Accuracy
nm Expected %T Tol. ±%T Meas. %T Result
1 590.0 10.1 ± 1.2 10.1 Pass
2 590.0 9.4 ± 1.2 9.4 Pass
Place Standards 1 - 2 in positions 1 - 2
Press Start Test
or
Press ESC to save test
Change to Abs
Add Std
Delete Std
Start Test

If the test fails follow these guidelines:

- Repeat the test twice to verify that the test is failing consistently.
- Make sure you follow the guidelines provided with the standard reference materials.
- Contact technical support for more troubleshooting advice.

Noise Measurement

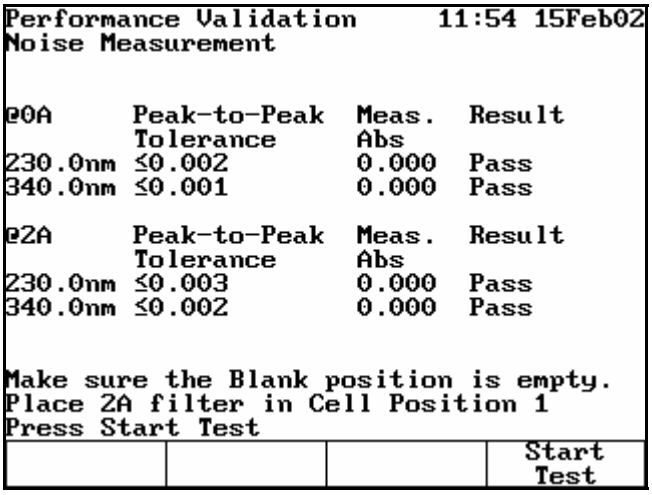
This test measures the amount of noise (nms) at 260 nm.

All test parameters are determined by instrument specifications and cannot be changed by the user. When running the noise test, remember that:

- The OA measurement should be performed with the cell holder empty. The 2A measurement should be performed with a 2A filter.

1. **Highlight Noise Measurement.**

2. **Press *Enter*.**



3. **Make sure that the Blank position is empty and insert the 2A filter in position #1.**

   The test results at 2A should be ignored if you do not have a filter installed in position #1.

4. **Press *Start Test* to run the test.**

   The results of the test appear on the screen, indicating pass or fail for each wavelength.

If the test fails follow these guidelines:

- Repeat the test twice to verify that the test is failing consistently.
- Make sure the instrument is warmed up for at least 30 minutes, with the standby mode feature turned off.
- Make sure the instrument is plugged into a stable power supply.
- Contact technical support for more troubleshooting advice.

Stray Light

This test measures the amount of stray light at selected wavelengths and compares them to expected values. The wavelengths and expected values are preset and cannot be changed. Running the stray light test takes approximately 30 seconds.

When running the stray light test remember that:

- You will need Stray Light standards that are designed to measure stray light at 198 nm, 220 nm, 340 nm, (i.e., must have ≤0.1%T at the wavelength of interest.
- Position B should be empty.
- Position #1 should be used for the 198 nm stray light standard (SRE 198 or equivalent).
- Position #2 should be used for the 220 nm stray light standard (SRE 220 or equivalent).
- Position #3 should be used for the 340 nm stray light standard (SRE 340 or equivalent).

1. **Highlight Stray Light.**
2. **Press *Enter*.**

Running the test

With the Stray Light screen displayed, make sure the wavelengths and tolerances are set correctly.

1. **Press *Start Test* to run the test.**

   The results of the test appear on the screen, indicating pass or fail for each wavelength.

If the test fails follow these guidelines:

- Repeat the test twice to verify that the test is failing consistently.
- Make sure all the filters being used are specifically designed to measure stray light at the specific wavelengths.
- Verify that the filters are placed in the correct cell positions.
- Contact technical support for more troubleshooting advice.

Internal Printer test

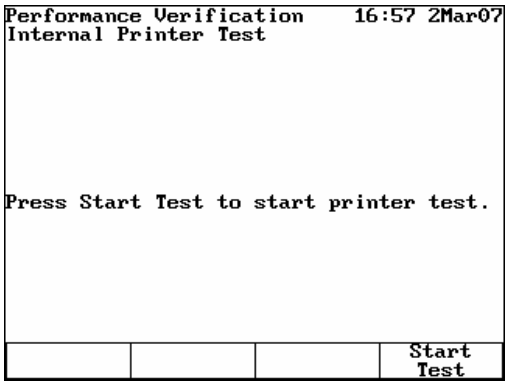
The Internal Printer test allows you to verify that the internal printer is functional. To run the test, you will need to have an internal printer installed. Running the internal printer test takes no more than 20 seconds after you press Stop.

1. **From the Utility screen, verify that the internal printer is installed properly and is selected.**

   If necessary, press *Utility* and then select the internal printer.

2. **With the Performance Verification screen displayed, highlight Internal Printer Test.**

3. **.Press *Enter*.**



4. **Press *Start Test* to run the test.**

   The test print routine appears on the printed results.

   While the test is running, you can press *Stop Test* to stop it.

If the test fails follow these guidelines:

- Make sure that the internal printer is the selected as the printer device on the Utility screen.
- Make sure that the internal printer is installed correctly. (Return to the main screen and press *Enter*. If the paper does not move, the printer may not be installed correctly.)
- Make sure that the thermal paper is threaded with the thermal side toward the printer head (the outside surface of the roll is the thermal surface).
- Contact technical support for more troubleshooting advice.

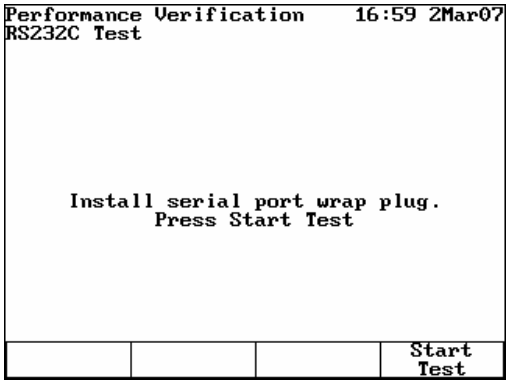
RS232 test

The RS232C test allows you to verify that the RS232C port is functional. To run the test, you will need to have the RS232 Test Plug accessory attached to the RS232C port on the back panel of the instrument. Running the RS232C test takes approximately five minutes.

1. **Verify that the RS232 Test Plug is securely attached to the RS232C port on the back panel of the instrument.**

2. **With the Performance Verification screen displayed, highlight RS232C Test.**

3. **Press *Enter*.**



4. **Press *Start Test* to run the test.**

   When the test is complete, the message "RS232 OK" or RS232 Failed" appears on the screen.

   While the test is running, you can press *Stop Test* to stop it.

If the test fails follow these guidelines:

- Make sure that the test plug is installed correctly and is not loose or damaged.

- Contact technical support for more troubleshooting advice.

Bio Tests

The Evolution 60 instrument allows you to run a variety of popular assays frequently used to characterize samples in the life science laboratory. These tests fall into the following categories:

- Nucleic acid measurements
- Protein measurements
- Cell growth analysis
- Oligonucleotide calculations

All of the parameters for the Bio Tests described in this chapter are set at the factory. If you want to change the parameters, specify a different name for the test and save the new test parameters.

To get started, press *Test*. When the Test Types screen appears, highlight Bio Tests and press *Enter.*

The list of Bio Test categories appears.

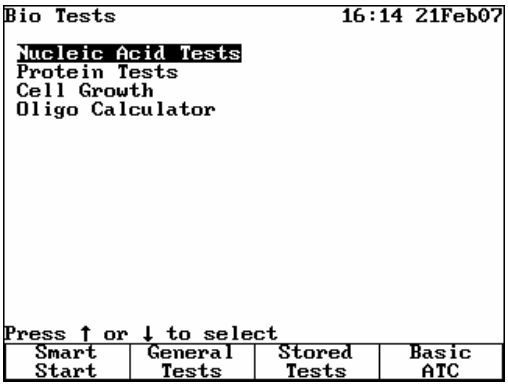


Table of parameters

Parameters used for each test and the literature reference for each test is located in the Parameters chapter.

You can use this list as a reference when you are setting up tests.

SmartStart feature

The SmartStart feature enables you to select the test methods you use most frequently and have them appear when you start up your instrument. If your laboratory runs only a single test, use the SmartStart feature to select it and it will appear each time you start up your instrument. Similarly, if you have a set of tests you run, you can use SmartStart to select them so the list appears when you start up the instrument.

Setting up a single-test SmartStart

1. **With the Bio Tests screen displayed, press *Stored Tests*.**

   A list of all the tests on the instrument appears on the screen.

2. **Scroll down through the list until the appropriate test is highlighted.**

3. **When the appropriate test is highlighted, press *Select Test* to add the selected test to the SmartStart menu.**

   An arrow sign ">" will indicate the test has been selected.

4. **Press *Load Test*.**

   The parameter screen of the test you selected will be displayed.

Note At this point, you can power down the instrument and then power it back up. When it starts up again, the parameter screen for the selected test will be displayed. ▲

Setting up a multiple-test SmartStart

1. **Press *Stored Tests*.**
2. **Scroll through the list until the first appropriate test is highlighted.**
3. **Press *Select Tests* to add the selected test to the SmartStart menu.**
4. **Continue scrolling through the list and adding tests until all the appropriate selections are made.**
5. **Press *Esc* until you return to the Bio Tests screen.**

Note At this point, you can power down the instrument and then power it back up. When it starts up again, the list of tests you've selected will be displayed. ▲

Nucleic acid measurement

You can use these tests to determine the concentration and purity of nucleic acid in a given sample.

DNA Direct 260 – Measures the DNA concentration by direct absorbance at 260 nm. An optional background correction at 320 nm is available

DNA 260/280 - Measures absorbance at 260 and 280 nm; determines DNA concentration and purity based on absorbance ratio and absorbance difference. An optional protein concentration calculation and background correction at 320 nm is available..

DNA 260/280 with scan – Records absorbance using a scan between 225 and 325 nm; determines DNA concentration and purity based on absorbance difference. An optional protein concentration calculation background correction at 320 nm is available.

DNA 260/230 – Measures absorbance at 260 and 230 nm; determines DNA concentration and purity based on absorbance ratio and absorbance difference. An optional protein concentration calculation and background correction at 320 nm is available.

DNA 260/230 with scan – Records absorbance using a scan between 225 and 325 nm; determines DNA concentration and purity based on absorbance ratio and absorbance difference. An optional protein concentration calculation and background correction at 320 nm is available.

dsDNA - Measures absorbance at 260 nm; calculates concentration based on absorbance and concentration factor.

ssDNA, RNA – Measures absorbance at 260 nm; calculates concentration based on absorbance and concentration factor.

Oligonucleotides (Oligos) – Measures absorbance at 260 nm; calculates concentration based on absorbance and concentration factor or calculates concentration based on absorbance and concentration factor determined by the oligo calculator.

Several of these categories include multiple tests that are similar, so the section may not include screen samples for each test. For example, the parameters are the same for the Direct UV measurement of ssDNA and RNA tests, but the factor used to convert absorbance to concentration is different. Similarly, for the Direct UV measurement of oligonucleotides tests, the parameters are also the same, but the factors used to convert absorbance to concentration are different. For a complete list of all parameters and calculations for each test, refer to Parameters or Calculations for Software, Calculations for Bio Tests Software, Calculations for Oligo Calculator.

DNA (260/280) and DNA (260/230)

These tests function almost identically - the only difference is the wavelengths used for the measurements. One test measures absorbance at 260 and 280 nm, while the other measures absorbance at 260 and 230 nm.

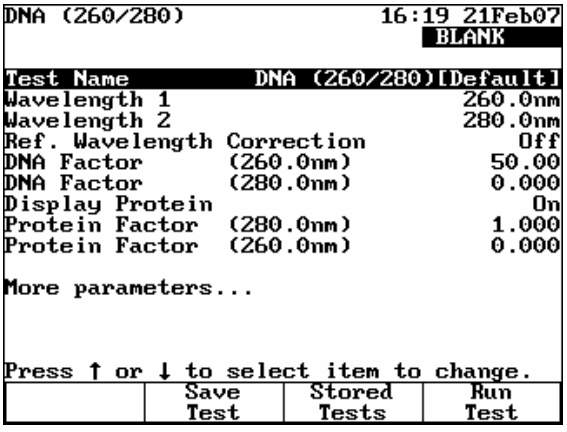
The measurement of DNA purity using the 260/230 ratio is commonly employed when Phenol is used in the process of extracting DNA. Phenol has a high absorbance at 230 nm and is easily detected using this test method.

Refer to the Parameters chapter for a description of the parameters and the Calculations for Bio Tests Software chapter for the default values

To get started, with the Bio Tests screen displayed, highlight Nucleic Acid Tests and press *Enter*. A list of nucleic acid test appears. Highlight DNA (260/280).

Note The following screens show the parameters for the DNA (260/280) test. For the DNA (260/230) test, Wavelength 2 is set to 230 nm. ▲

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲



Setting up test parameters

With the DNA (260/280) or DNA (260/230) screen displayed, highlight the name of the parameter you want to set.

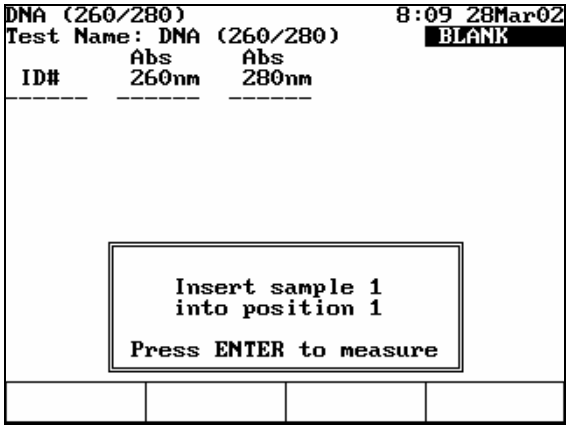
Measuring Protein Concentration

To measure the protein concentration using this test method, select Measure Protein and press *Enter*.

When the parameters are set, press *Save Test* to save the test under a new name or *Run Test* to measure the blank and unknowns.

Measuring unknowns

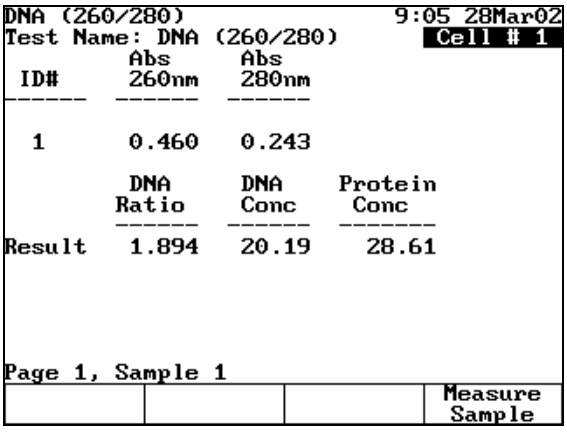
Measuring unknowns automatically (using Auto 6 or Auto 3)


DNA (260/280) 8:09 28Mar02
Test Name: DNA (260/280) BLANK
ID# Abs 260nm Abs 280nm
Insert sample 1
into position 1
Press ENTER to measure

1. **Place the blank and unknowns in the correct cell positions.**

2. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the sample measurements on the screen.



Measuring unknowns manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Test*.**

2. **Place the blank and unknown in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

DNA with Scan (260/280) and DNA with Scan 260/230)

The DNA measurement with scan tests include two tests that function almost identically - the only difference is in the wavelengths used for the measurements. In both cases the scan is measured from 225 to 325 nm. One test measures absorbance at 260 and 280 nm, while the other measures absorbance at 260 and 230 nm.

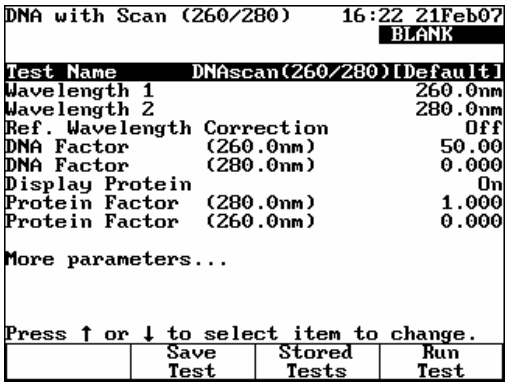
The measurement of DNA purity using the 260/230 ratio is commonly employed when Phenol is used in the process of extracting DNA. Phenol has a high absorbance at 230 nm and is easily detected using this test method.

Refer to the Parameters chapter for a description of the parameters and the Calculations for Bio Tests Software chapter for the default values

To get started, with the Bio Tests screen displayed, highlight Nucleic Acid Tests and press *Enter*. A list of nucleic acid test appears. Highlight DNA with Scan (260/280). The DNA with Scan (260/280) parameter screen appears.

Note The following screens show the parameters for the DNA (260/280) test. For the DNA (260/230) test, Wavelength 2 is set to 230 nm. ▲

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲



Setting up test parameters

With the DNA with Scan (260/280) or DNA with Scan (260/230) screen displayed, highlight the name of the parameter you want to set.

Measuring Protein Concentration

To measure the protein concentration using this test method, select Measure Protein and press *Enter*.

When the parameters are set, press *Save Test* to save the test under a new name or *Run Test* to measure the blank and unknowns.

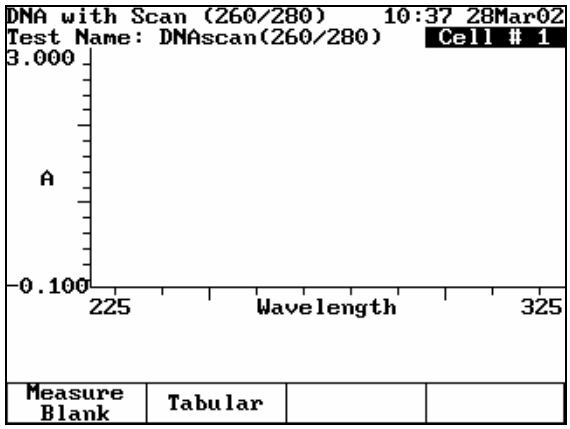
Collecting a baseline scan

With the DNA with Scan (260/280) or DNA with Scan (260/230) screen displayed, follow these steps to collect a baseline scan.

Note If a 6-Position Cell Holder is installed, be sure to place the blank in the B position. The instrument always uses the B position to collect the baseline. ▲

1. **Press *Run Test*.**

   The DNA with Scan measurement screen appears.



2. **Place the blank in the B position.**

3. **Press *Measure Blank* to collect the baseline.**

   When the instrument is finished measuring the blank, the message disappears.

Note If you want to switch between tabular and graphical displays, press *Graph* or *Tabular*. ▲

Measuring the sample

With the DNA with Scan (260/280) or DNA with Scan (260/230) screen displayed, follow these steps to measure the sample.

If a 6-Position Cell Holder is installed, be sure to place the unknown in cell position #1.

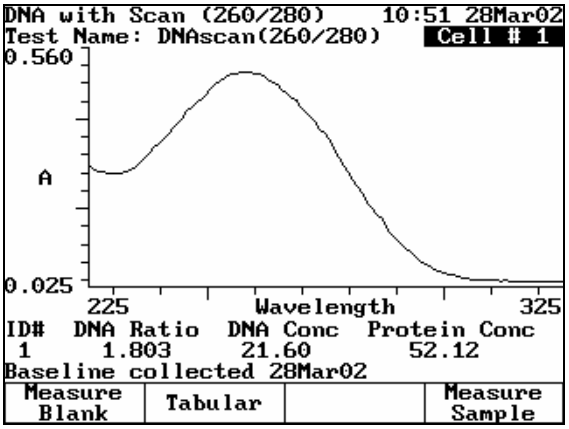
Note The instrument always uses cell position #1 to measure the sample. ▲

1. **Press *Measure Sample* to measure the sample.**

   When the instrument is finished measuring the absorbance scan, it displays a graph of the scan along with the sample ID#, DNA ratio, DNA concentration and protein concentration.

Note If you want to switch between tabular and graphical displays, press *Graph* or *Tabular*. ▲

Note You may need to use the up and down arrow keys to view all the data for the screen. ▲



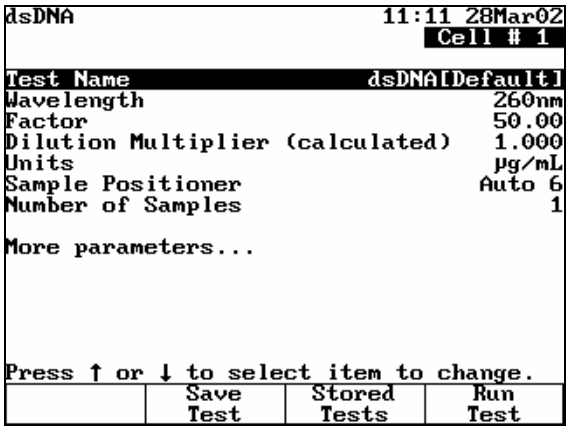
DNA Direct 260, dsDNA, ssDNA, RNA and Oligos (entered factor) measurements

These test measurements are all set up and run using the same types of test parameters. Refer to the Parameters chapter for a description of the parameters and the Calculations for Bio Tests Software chapter for the default values.

To get started, highlight Nucleic Acid Tests and press *Enter*. A list of nucleic acid test appears. Highlight the desired test and press *Enter*. The dsDNA, ssDNA, RNA or Oligos (entered factor) parameter screen appears.

Note The following screens show the parameters for the dsDNA test. ▲

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

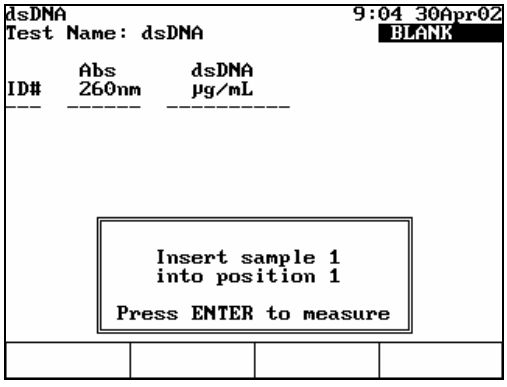


Setting up test parameters

1. **Highlight the name of the parameter you want to set.**
2. **When the parameters are set, press *Save Test* to save the test under a new name or *Run Test* to measure the blank and unknowns.**

Measuring unknowns

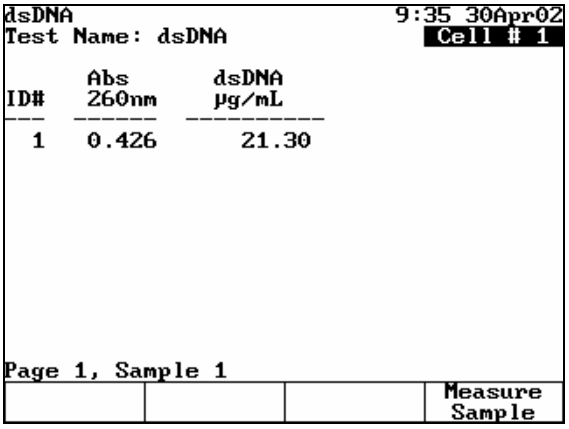
Measuring unknowns automatically (using Auto 6 or Auto 3)



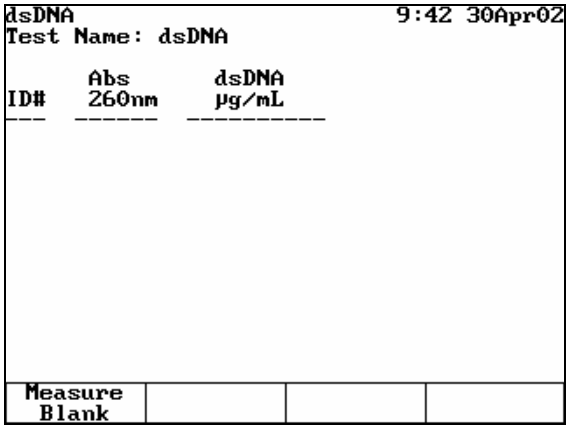
1. **Place the blank and unknowns in the correct cell positions.**

2. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the sample measurements on the screen.



Measuring unknowns manually (using Manual 6 or Single Cell Holder)



1. **Press *Run Test*.**

2. **Place the blank and unknown in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

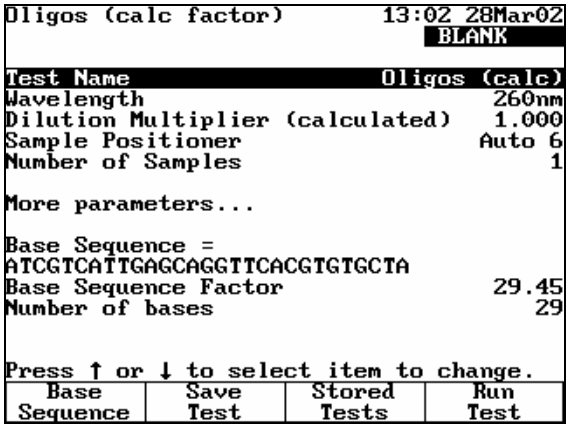
Oligos (calculated factor)

The Oligos (calculated factor) measurement calculates molecular weight, extinction coefficient and a concentration factor for a short (< 40-mer) oligonucleotide based on a base sequence that you enter manually.

This concentration factor is used to calculate the concentration of oligonucleotides in your sample from the absorbance measurement. Refer to the Parameters chapter for a description of the parameters and the Calculations for Oligo Calculator chapter, for the default values.

To get started, highlight Nucleic Acid Tests and press *Enter*. A list of nucleic acid test appears. Highlight the desired test and press *Enter*. The Oligos (calc factor) parameter screen appears.

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲



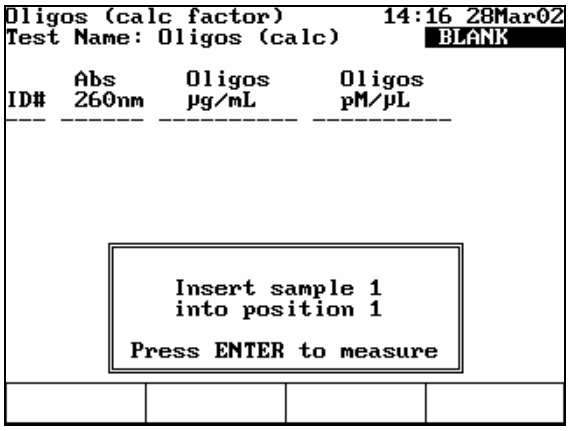
Setting up parameters

1. **Highlight the name of the parameter you want to set.**

2. **When the parameters are set, press *Save Test* to save the test or *Run Test* to measure the blank and unknowns.**

Measuring unknowns

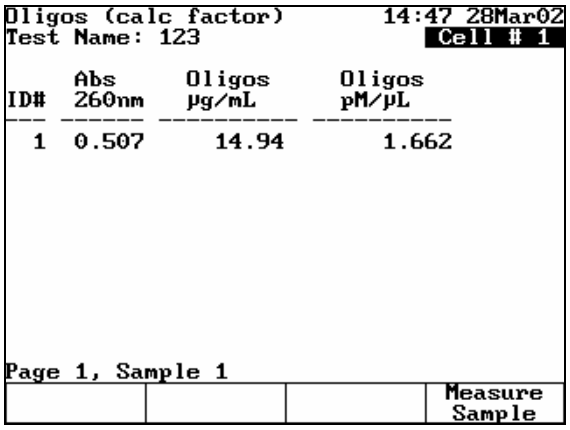
Measuring unknowns automatically (using Auto 6 or Auto 3)


Oligos (calc factor) 14:16 28Mar02
Test Name: Oligos (calc) BLANK
ID# Abs 260nm Oligos µg/mL Oligos pM/µL
Insert sample 1
into position 1
Press ENTER to measure

1. **Place the blank and unknowns in the correct cell positions.**

2. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays absorbance, oligo concentration in µg/mL and pmol/µL.


Oligos (calc factor) 14:47 28Mar02
Test Name: 123 Cell # 1
ID# Abs 260nm Oligos µg/mL Oligos pM/µL
1 0.507 14.94 1.662
Page 1, Sample 1
Measure Sample

Measuring unknowns manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Test*.**

2. **Place the blank and unknown in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

Protein measurements

Use the following test methods to determine the concentration of protein in a given sample:

Protein Direct 280 – Measures the absorbance of the sample at 280 nm and determines the protein concentration based on a factor. An optional correction at 320 nm is available.

Bradford – Measures absorbance at 595 nm; determines concentration for either standard or micro sample concentrations.

Lowry – Measures absorbance at 550 nm for standard and 770 nm for micro; determines concentration for either standard or micro sample concentrations.

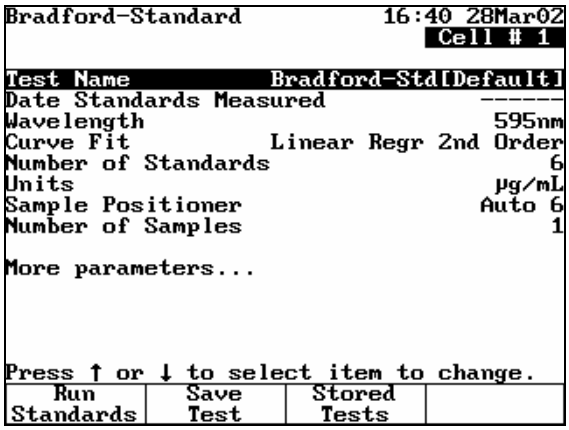
Bicinchoninic Acid (BCA) – Measures absorbance at 562 nm; determines concentration for either standard or micro sample concentrations.

Biuret – Measures absorbance at 540 nm.

Several of these categories include multiple tests that are similar, so this section includes screen samples for the standard Bradford test only. For a complete list of all parameters for each test, refer to the Parameters chapter for a list of calculations used for the tests, refer to the Calculations for Bio Tests Software chapter for default values.

To get started, with the Bio Tests screen displayed, highlight Protein Tests and press *Enter*. A list of protein tests appears. Highlight the desired test and press *Enter*. The Bradford-Standard parameter screen appears.

Note The following screens show the parameters for the **Bradford-Standard** test. All the other protein standard curve methods work in a similar fashion. ▲



Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Setting up test parameters for a standard curve

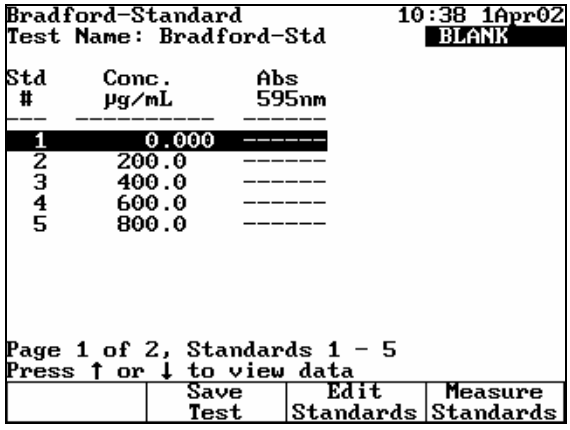
1. **Highlight the name of the parameter you want to set**

   Set the parameters for measuring the standards. Refer to the list of parameters in the Parameters chapter for a description of the parameters and Calculations for Bio Tests Software for the default values.

2. **When the parameters are set, press *Save Test* to save the test or *Run Standards* to measure the blank and standards.**

3. **If you need to edit concentration values, select the standard you want to edit and press *Edit Standards*.**

   From the Edit Standards window you can edit, add or delete one or all standards



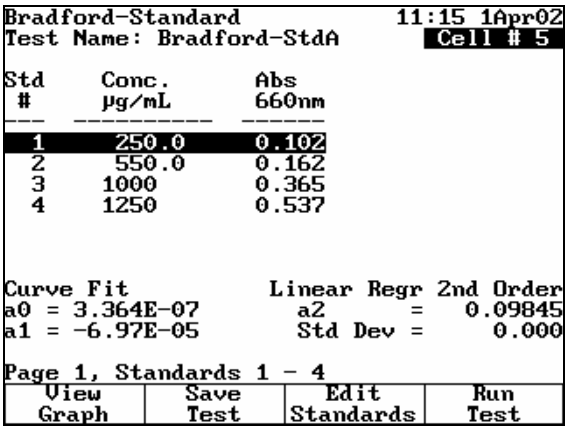
Measuring the standards for a standard curve

Measuring standards automatically (using Auto 6 or Auto 3)

1. **Place the blank and standards in the correct cell positions.**

2. **When all the standards are correct, press *Measure Standards* to set up and run the standards.**

The instrument automatically measures the blank first and then measures the standards. When the instrument has measured all the standards, the Standards screen appears, showing the absorbance of each standard, along with the slope, intercept and correlation coefficient of the standard curve.


Bradford-Standard 11:15 1Apr02
Test Name: Bradford-StdA Cell # 5
Std # Conc. µg/mL Abs 660nm
1 250.0 0.102
2 550.0 0.162
3 1000 0.365
4 1250 0.537
Curve Fit Linear Regr 2nd Order
a0 = 3.364E-07 a2 = 0.09845
a1 = -6.97E-05 Std Dev = 0.000
Page 1, Standards 1 - 4
View Graph
Save Test
Edit Standards
Run Test

Measuring standards manually (using Manual 6 or Single Cell Holder)

1. **Place the blank and standards in the correct cell positions.**

2. **When all the standards are correct, press *Measure Standards* to set up and run the standards.**

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Standards* to measure the standards.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the standards manually.

When the instrument has measured all the standards, the Standards screen appears, showing the absorbance of each standard, along with the slope, intercept and correlation coefficient of the standard curve.

You can use this screen to:

- Edit the standards (press *Edit Standards*)
- Display a graph of the standard curve data (press *View Graph*)
- Save the standard curve (press *Save Test*)
- Measure your samples (press *Run Test*)

Measuring protein samples

Measuring standards automatically (using Auto 6 or Auto 3)

With the Standards screen displayed, place the blank and unknowns in the correct cell positions.

1. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the absorbance and concentration of each unknown.

Measuring standards manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Test*.**

2. **Place the blank and unknown in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

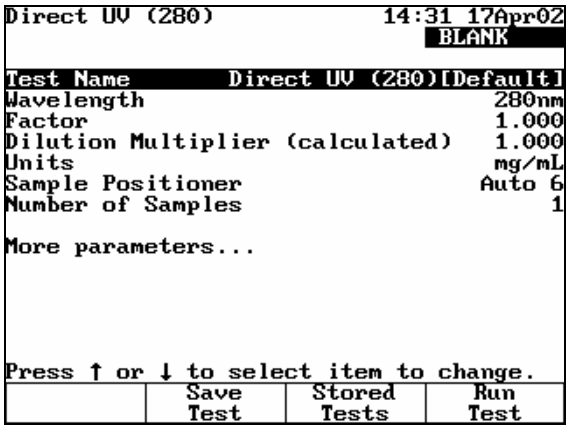
Direct UV (280) and Direct UV (205)

The Direct UV methods determine protein concentration based on absorbance at either 280 or 205 nm. Refer to the Parameters chapter for a description of the parameters and Calculations for Bio Tests Software chapter for the default values.

To get started, highlight Protein Tests and press *Enter*. Then highlight Direct UV (280) and press *Enter*. The Direct UV (280) parameter screen appears.

Note The following screens show the parameters for the Direct UV tests at 280 nm. For the Direct UV test at 205 nm, the wavelength is set to 205 nm. ▲

Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲



Setting up test parameters

1. **Highlight the name of the parameter you want to set.**
2. **When the parameters are set, press *Save Test* to save the test or *Run Test* to measure the blank and unknowns.**

Measuring the sample

Measuring standards automatically (using Auto 6 or Auto 3)

1. **Place the blank and unknowns in the correct cell positions.**
2. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the absorbance and concentration of each unknown.

Measuring standards manually (using Manual 6 or Single Cell Holder)

1. **Press *Run Test*.**
2. **Place the blank and unknown in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

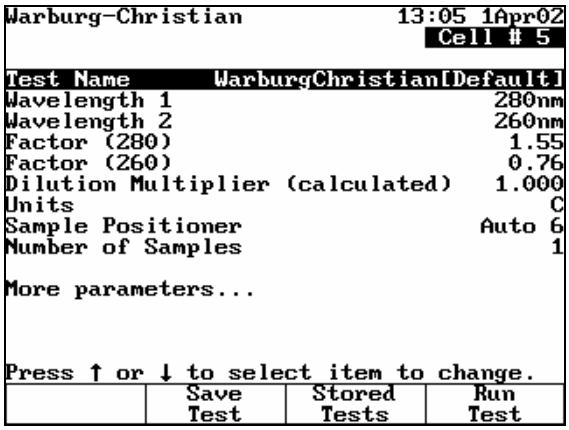
4. **Press *Measure Sample* to start the measurement.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

Warburg-Christian

The Warburg-Christian analysis uses an absorbance difference measurement at 280 and 260 nm to determine the protein concentration of an unknown. Refer to the Parameters chapter for a description of the parameters and Calculations for Bio Tests Software chapter for the default values.

To get started, highlight Protein Tests and press *Enter*. Then highlight Warburg-Christian and press *Enter*. The Warburg-Christian parameter screen appears.



Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Setting up the test parameters

1. **Highlight the name of the parameter you want to set.**
2. **When the parameters are set, press *Save Test* to save the test or *Run Test* to measure the blank and unknowns.**

Measuring the sample

Measuring unknowns automatically (using Manual 6 or Single Cell Positioner)

1. **Place the blank and unknowns in the correct cell positions.**

2. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the absorbance and concentration of each unknown.

Measuring standards manually (using Manual 6 or Single Cell Positioner)

1. **Press *Run Test*.**

2. **Place the blank and unknown in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

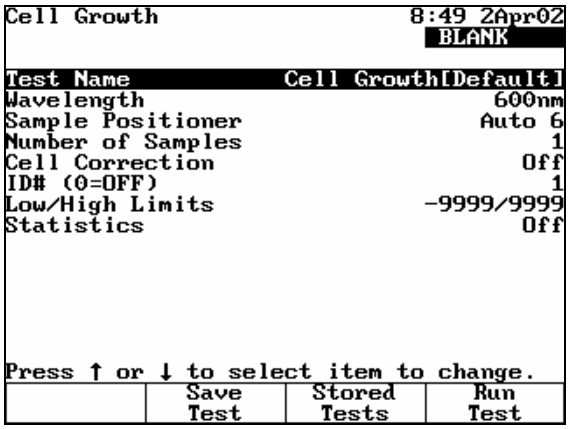
   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

   When the instrument has measured all the unknowns, it displays the absorbance and concentration.

Cell growth

The cell growth measurement uses absorbance at 600 nm to indicate the progress of cell growth in a sample. The instrument does not perform any calculations or graphing for the data.

To get started, highlight Cell Growth and press *Enter*. The Cell Growth setup screen appears.



Note If Cell Correction is ON, you must run the Setup Correction program before you can access *Run Test* or *Measure Samples*. ▲

Setting up test parameters

Highlight the name of the parameter you want to set.

Using the Correction Factor

If you are using different spectrophotometers in your laboratory, particularly if they are from a different manufacturer, you may notice that readings for cell growth vary widely between these instruments. This occurs because some instruments use different types of optical systems and detection schemes to collect data.

If you are using different spectrophotometers in your lab and would like to compare data from a different instrument, use the correction factor option.

The correction factor allows you to scale the readings from your Evolution 60 to match another spectrophotometer in your lab. Simply measure the difference between the two instruments and use the factor to adjust for this difference. For more information, contact your sales representative

When the parameters are set, press *Save Test* to save the test or *Run Test* to measure the blank and unknowns.

Measuring the sample

Measuring unknowns automatically (using Manual 6 or Single Cell Positioner)

1. **Place the blank and unknowns in the correct cell positions.**

2. **Press *Enter* to start the measurements.**

   The instrument automatically measures the blank first, then measures the unknowns and displays the absorbance and concentration of each unknown.

Measuring standards manually (using Manual 6 or Single Cell Positioner)

1. **Press *Run Test*.**

2. **Place the blank and unknown in the correct cell positions.**

   If a 6-Position Cell Holder is installed, you can place up to five samples in the cell holder.

3. **Press *Measure Blank* to measure the blank.**

   If a 6-Position Cell Holder is installed, it automatically moves to the B position to measure the blank. When it completes the measurement, it returns to its previous cell position.

4. **Press *Measure Sample* to start the measurement.**

   If a 6-Position Cell Holder is installed, press the cell position buttons to re-position the cell holder and measure the rest of the unknowns manually.

When the instrument has measured all the unknowns, it displays the absorbance and concentration.

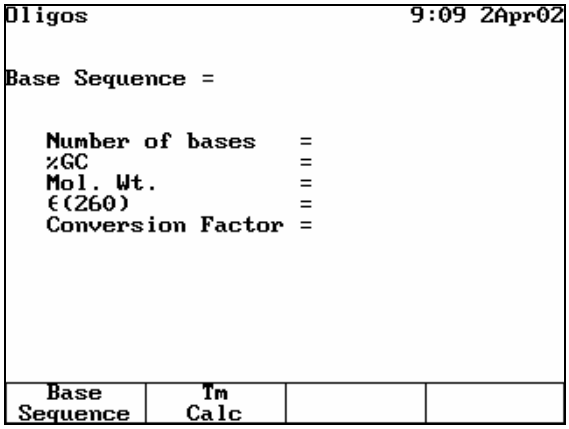
Oligo calculator

The oligonucleotide calculator determines the following data for a base sequence that you enter.

- Number of bases
- Percent GC content
- Molecular weight
- Molar Absorptivity or Extinction Coefficient (ε)
- Conversion factor to convert nucleotide absorbance at 260 nm concentration
- T_m for oligos of up to 40 bases for DNA-DNA, DNA-RNA and RNA-RNA hybrids

Refer to the Parameters chapter for a description of the parameters and the Calculations for Oligo Calculator chapter for the default values.

To get started, highlight Oligo Calculator and press *Enter*. The Oligos screen appears.



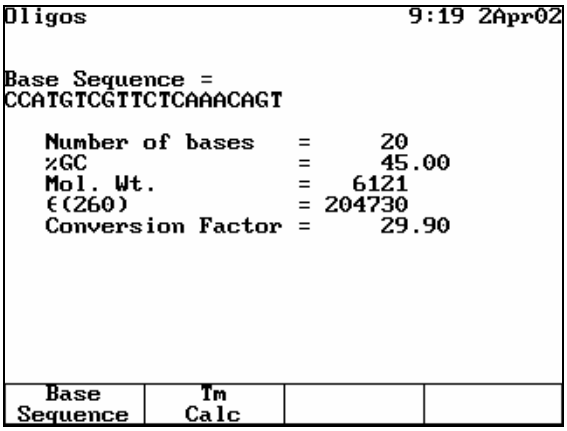
Using the oligo calculator

1. **Press *Base Sequence*.**
2. **Select the appropriate character for the base you want to enter.**

3. **Press *Add Base* to add the base to the sequence.**

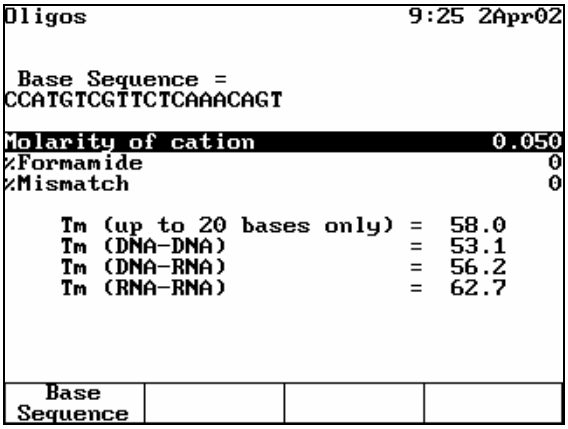
4. **When the base sequence is correct, press *Accept Sequence* to accept it.**

   The instrument calculates and displays the results.



5. **To determine the theoretical T_m of the sequence, press T_m Calc.**

   The T_m calculation screen appears.

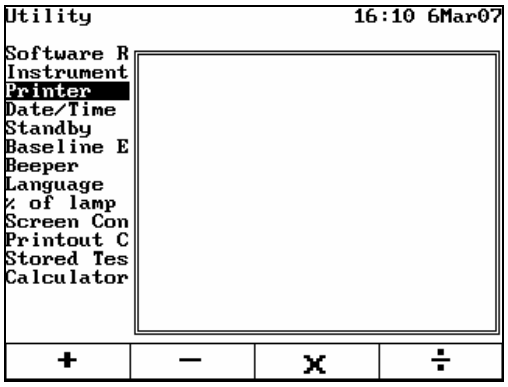


6. **Enter the % formamide and % mismatch (if known) that will be used to calculate the T_m.**

   The calculated T_m values are shown on the screen.

Utility Calculator Function

1. **From the Utility menu highlight *Calculator* and press *Enter*.**

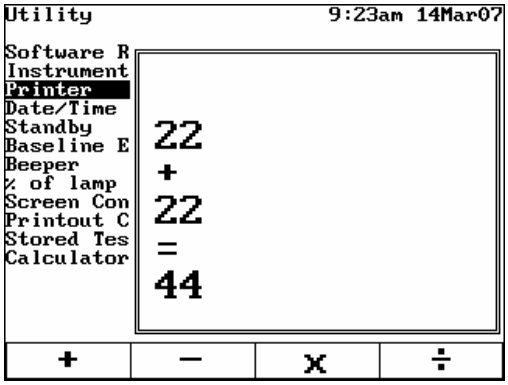


2. **Use the numeric keypad to enter the desired value.**

3. **Press the desired function (+, -, *x or* ÷).**

4. **Enter the second desired value and press *Enter* to calculate the total.**

Note You can only add, subtract, multiply, or divide two lines of numbers at a time. ▲



Maintenance

The spectrophotometer is durable and reliable, so routine maintenance is minimal. This section includes complete instructions for:

- Routine care, cleaning and maintenance of the instrument and cells.
- Changing the fuse and voltage setting.

⚠ Warning Operating the instrument with the cover off exposes the operator to potentially dangerous voltages and ultraviolet (UV) radiation. Therefore, we recommend that only authorized service representatives perform procedures requiring removal of the instrument cover and replacement of electrical components. To protect both yourself and the instrument, be sure to contact an authorized service representative to perform any service procedure you do not feel comfortable performing. ▲

Routine care

Routine care for the spectrophotometer does not require a lot of time. To help minimize maintenance time and to increase the life and performance of your instrument, please follow these guidelines:

- Always replace the dust cover when the instrument is not turned on to prevent dust from accumulating in and on the instrument.
- Do not use or store the instrument in a corrosive environment.
- Gently wipe the outside of the instrument, including the keypad, with a soft cloth to remove any dust or

spills. Water, isopropyl alcohol and other common laboratory cleaning agents may be used if necessary.

Always clean up spills as soon as they occur to prevent or minimize damage to the instrument. If concentrated acids or bases, or any hydrocarbon materials, are spilled on the instrument, be sure to clean up the affected area immediately.

Cleaning and maintaining cells

Carefully check the condition of the cuvettes and other cells used to measure samples. If they are chipped, cracked or scratched it is important to discard the damaged cell(s) and replace them with new ones.

Ensuring your cuvettes are clean both inside and out is important to the quality of your results for two reasons: 1) contaminating material may absorb light resulting in falsely high absorbance readings; and 2) contaminants in the cell may react chemically with subsequent reagents or standards introduced into the cell.

Cleaning methods depend to some extent on the nature of the contaminating material. It is important to identify the residual material in the cell that needs to be removed. Refer to the following table for suggestions on cleaning methods, solvents and material.

Solvent	Examples	Suggested Cleaning Methods
Aqueous	Protein, Biologics, DNA	• Warm water with detergent • Dilute Nitric Acid (<10%) rinse • Copious water rinse
Aqueous	Salt solutions	• Dilute Nitric Acid (<10%) rinse • Copious water rinse

Solvent	Examples	Suggested Cleaning Methods
Aqueous	Basic solutions	• Warm water with detergent • Dilute Nitric Acid (<10%) rinse • Copious water rinse
Organic	Hydrocarbons, small molecules, oils	• Rinse with organic solvent • Warm water with detergent • Dilute Nitric Acid (<10%) rinse • Copious water rinse
Organic	Alcohol solutions	• Rinse with similar alcohol, acetone, or other solvent • Copious water rinse
Organic	Acidic solutions	• Rinse with organic solvent • Warm water with detergent • Dilute Nitric Acid (<10%) rinse • Copious water rinse
Organic	Basic solutions	• Rinse with organic solvent • Warm water with detergent • Dilute Nitric Acid (<10%) rinse • Copious water rinse

Notice Keeping the cell clean is very important for a long cell life. ▲

- Never store cuvettes long term in a water or solvent bath between uses. If the solvent you are using dries impurities in the water or solvent be deposited on the inside of the cell, causing permanent damage.

- Use only lens cleaning tissue/paper or fine soft cloth to wipe optical surfaces. Most paper products (such as facial tissues, paper towels, etc.) contain wood fibers that can damage the cell material.

- At the end of the day, ensure that all cells are well cleaned and stored in a suitable container after drying.

Term	Definition
Dilute acid	Dilute Nitric Acid (<10%)
Acid	Hydrochloric (5M) acid or Nitric Acid (5M) (see Note below)
Solvent rinse	Rinse with the solvent that was originally used solvated your analyte
Copious water rinse	Use a pure water (e.g., deionized, distilled, RO) and rinse at least 10 times
Detergent	Use a neutral pH detergent (Triton X-100), if available, to dilute acid wash; water rinse to remove residue

Note Do not use 5M Nitric Acid on an Anti-reflection mirror coated cells. ▲

Notice It is not recommended that you use an ultrasonic cleaning baths for your cells. Each bath generates a different frequency; therefore, if your bath operates at the resonant frequency of a cell, the cell will break. If a cell was cleaned in an ultrasonic bath, the warranty is void by the manufacturer. ▲

Notice Do not dry cells in an oven. ▲

Micro flowcells can be kept clean by:

- Flushing well with a solvent after use.

- Aspirating dilute acid, base, non-filming detergent or Clorox through the cell in short bursts.
- Storing with distilled water in the cell.

Cleaning the windows of the sample compartment

Follow these guidelines to clean the windows of the sample compartment:

- **Do not use acetone or abrasive materials** to clean the windows of the sample compartment. Instead, use a non-abrasive laboratory cleaning solution (such as a commercial cell cleaning solution), distilled water or alcohol.
- Use the liquid and a soft, lint-free cloth to clean the windows. Do not apply too much pressure or the surface of the windows may be damaged. Be sure to remove all fingerprints.

Changing the fuse

The fuse is located in the power entry module located at the center of the back panel of the instrument.

- 120VAC, 2.5A, Slo-Blo
- 240VAC, 1.25A, Slo-Blo (2 required)

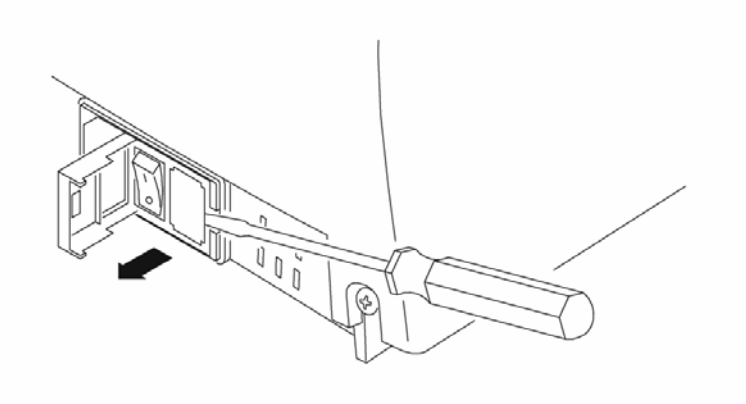
Notice The instrument fuse must be replaced with the same type and rate. ▲

Notice If the fuse fails repeatedly, it may indicate a serious problem with the instrument. Contact technical support as soon as possible. ▲

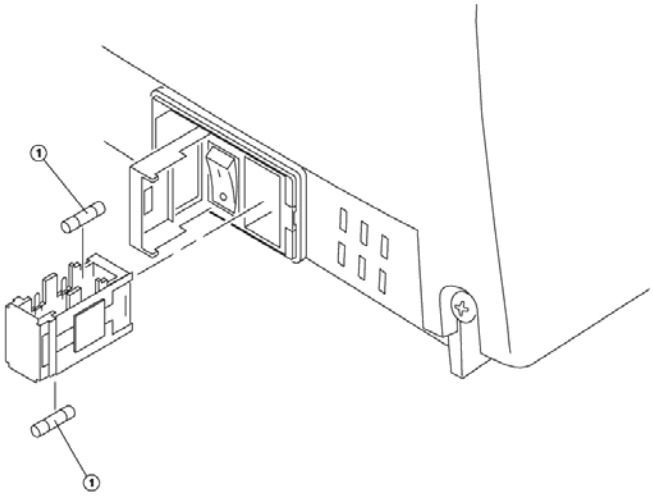
1. **Turn off and unplug the instrument from the wall outlet or power strip.**
2. **Position the instrument so you can access the power entry module on the back of the instrument.**

3. Remove the power cord.

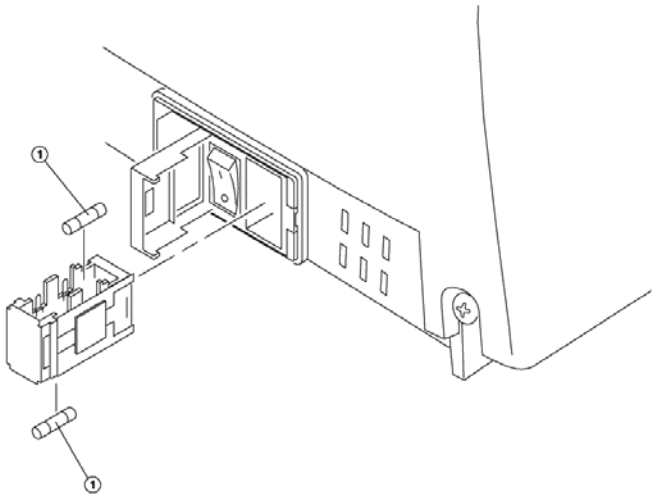
4. Insert a flat-blade screwdriver into the notch on the fuse cover and pry off the cover.



5. Use a flat-blade screwdriver to remove the fuse holder.



6. Unsnap both fuses to remove them.



7. **Insert the new fuses, pushing them in so they snap into place.**
8. **Replace the fuse cover.**
9. **Replace the power cord.**
10. **Plug the instrument back in to the wall outlet or power strip and turn on the power.**

Note If the fuse blows again, contact your distributor or technical support. ▲

Parameters

Parameter	Description
+ - x ÷	Enters math operators when in 'Calculator' mode (Utility)
% Formamide	Enter Percentage of formamide contained in the sample (Oligos tests)
% GC	Calculates Percentage of GC pairs contained in the sample (Oligos tests)
%Mismatch	Enter Mismatch value to calculate Tm (Oligos tests)
% of lamp life used	Displays estimated percentage of lamp life used (based on typical lamp life of five years) (Utility)
3-Pt Net	Enables you to calculate peak height from tangential baseline in graph (Scanning)
Absorbance	Enters absorbance value
Accept Name	Accepts the displayed Name entry (Test Name and Edit [Units])
Add Character	Adds highlighted character to Name entry (Test Name and Edit [Units])
Add nm	Allows you to add a wavelength and factor to the list in Multiwavelength tests and some Performance Verification tests
Area	Enables you to calculate area under peak in graph (Scanning)
AutoPrint	Turns automatic printout on or off
Autoscale	Rescales graph to original ranges of x and y axes (Kinetics, Scanning)
Base Sequence	Sequence of bases contained in the sample (Oligos – calc factor tests)
Baseline Expiration	Enters time when baseline for scan tests will need to be collected again (Utility)

Parameter	Description
Beeper	Turns audible signal for key presses on and off (Utility)
Calculation Baseline	Select the zero baseline or the tangential baseline to calculate area under the peak in graph (Scanning)
Calculator	Enables the calculator mode (Utility)
Cell Correction	Select option to automatically corrects for variances in absorption between the cuvettes (all test types)
Cell Position #	Displays position placed in light path (only with auto turret)
Change Mode Change to Abs Change to %T	Switches measurement modes (Basic A-%T-C and some Performance Verification tests)
Collect Baseline	Starts the collection of the baseline (Scanning only)
Concentration	Sets concentration value
Conc of Standard	Displays the entered concentration value (Adv A-%T-C)
Correction Mode	Select mode for cell correction (Discrete nms or Scan)
Cursor	Goes to Cursor tracking mode to view data points in graph (Kinetics, Scanning)
Cursor → ← Cursor	Moves cursor right or left on graph and displays data of each point (Kinetics, Scanning)
Curve Fit	Selects type of line fit calculation (Standard Curve tests)
Data File Name	Allows entry of name for data file when AutoSave = ON
Date Cell Correction	Displays date when cell correction data on cuvettes was last collected
Date Standards Measured	Displays date when standards were last measured with this instrument (Standard Curve tests)
Date/Time Setup	Enters current date and time settings for the instrument (Utility)

Parameter	Description
Delay Time	Enters time from Test Initiation to first measurement; allows for sample equilibration (ADV. A-%T-C and Kinetics)
Delete Character	Deletes last character of Name entry (Test Name and Edit [Units])
Delete File	Deletes test or data file from the Stored Tests Directory (Utility)
Delete Name	Deletes entire name to allow new entry (Test Name and Edit [Units])
Delete nm	Removes a wavelength and factor from the list (Multiwavelength and some Performance Verification tests)
Diluent Volume	Enters volume of diluent added prior to measurement (Dilution Multiplier in some Bio Tests)
Dilution Multiplier	Displays factor used to correct for sample dilution
Display Activity	Indicates whether results should include protein concentration
DNA ε(260)	Calculates extinction coefficient
DNA Factor	Enter factor to calculate DNA concentration (DNA Bio Tests)
Edit	Allows you to change a wavelength or factor in the list (Multiwavelength and some Performance Verification tests)
Edit Curve	Allows you to manipulate graph (Kinetics)
Edit Data	Enables you to select portion of the data in a table for recalculation of a result (Kinetics and Scanning)
Edit Graph	Allows you to manipulate graph (Scanning)
Edit Scale	Allows you to change graph axis scales and view individual data points (Scanning)
Factor	Enters factor to convert a datum to a result Abs x Factor 1 = Concentration Result Abs/min x Factor 2 = Kinetics Result Can be entered or calculated from concentration and absorbance values in ADV A-%T-C

<table>
<tr><th>Parameter</th><th>Description</th></tr>
<tr><td>Factor 1</td><td>Enters factor to convert a datum to a result

Abs(WL1) x Factor = Result
(Abs Ratio, Abs Diff, Multiwavelength tests)</td></tr>
<tr><td>Factor 2</td><td>Enters factor to convert a datum to a result

Abs(WL2) x Factor = Result
(Abs Ratio, Abs Diff, Multiwavelength tests)</td></tr>
<tr><td>Factor 3-31</td><td>Enters factor to convert a datum to a result

Abs (WL3-31) x Factor = Result
(Multiwavelength tests)</td></tr>
<tr><td>Graph</td><td>Displays graph of collected data
(Kinetics, Scanning)</td></tr>
<tr><td>ID #</td><td>Enter numeric identifier for measurement - autoincrements during test until turned off (set to 0)</td></tr>
<tr><td>Instrument Serial Number</td><td>Displays serial of the instrument
(Utility)</td></tr>
<tr><td>Intercept</td><td>Enter where line crosses the y-axis (Abs where conc=0)</td></tr>
<tr><td>Interval</td><td>Enter wavelength range between data points
(Scanning tests only)</td></tr>
<tr><td>Interval Time</td><td>Enter time between repeated readings
(Kinetics only)</td></tr>
<tr><td>Linearity Value</td><td>Enters a linearity value
(Kinetics only)

To help determine the linearity of the reaction during the measurement, the spectrophotometer offers a linearity parameter. This parameter is the difference between the changes in the absorbance of two measurements as shown in the following example:
<table>
<tr><th>Time</th><th>Abs</th><th>ΔA</th><th>Linearity</th></tr>
<tr><td>1</td><td>.1</td><td>---</td><td>---</td></tr>
<tr><td>2</td><td>.2</td><td>.1</td><td>---</td></tr>
<tr><td>3</td><td>.29</td><td>.09</td><td>P</td></tr>
<tr><td>4</td><td>.38</td><td>.09</td><td>P</td></tr>
<tr><td>5</td><td>.46</td><td>.08</td><td>P</td></tr>
<tr><td>6</td><td>.52</td><td>.06</td><td>F</td></tr>
</table>
Linearity is the ΔA between ΔA calculations

P = Pass and F = Fail</td></tr>
</table>

Parameter	Description
Load Test	Loads highlighted test from Stored Tests Directory into active memory and sets instrument to test parameters (Utility)
Lock/Unlock	Used to protect stored tests from accidental deletion or alteration; asks for password to allow user to lock or unlock the file (Utility)
Low/High Limits	Enter lowest and highest acceptable results, outside of which the result is flagged as 'Low' or 'High' (Adv. A-%T-C, Std Curve, Abs Ratio, Abs Diff, Kinetics, 3-Pt Net and some Bio Tests)
Math	Accesses manipulation functions of graph (Scanning)
Measure Blank (as function key)	Initiates measurement of blank
Measure Blank (as test parameter)	Selects frequency of zeroing the instrument Once or Every Reading (Kinetics)
Measurement Mode	Selects the type of photometric data reported for a measurement (Abs, %T, Conc) (in A-%T-C, Kinetics, Scanning, Multiwavelength)
Measure Samples	Initiates measurement of samples
Max, X	Enter a maximum x-value to manually rescale the graph (Kinetics, Scanning)
Max, Y	Enter a maximum y-value to manually rescale the graph (Kinetics, Scanning)
Min, X	Enter a minimum x-value to manually rescale the graph (Kinetics, Scanning)
	Enter a minimum y-value to manually rescale the graph (Kinetics, Scanning)
Molarity of cation	Enters molarity of Na* in incubation mixture (T_m calculation in Oligos tests)
Next Cursor	Selects a cursor point in functions using more than one cursor setting – Scan-Area and Scan-3-Pt Net calculations in graph (Scanning)
Number of bases	Enters number of bases in the oligonucleotide (Oligos tests)
Number of Matched Cuvettes	Enter number of cuvettes that will be run in the Correction Program (maximum of 5)

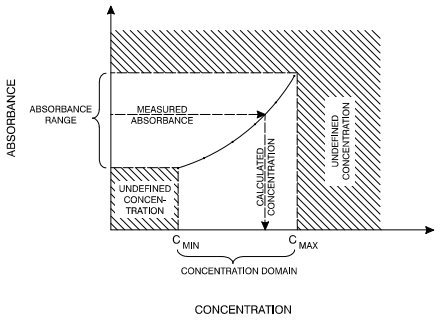
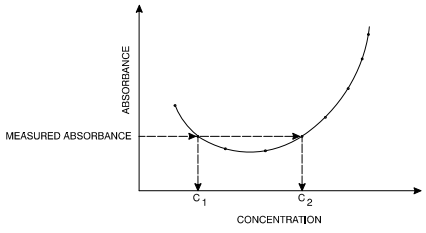
Parameter	Description
Number of Samples	Enter number of samples to be measured in the test (**Not** available in Kinetics or Scanning)
Number of Standards	Enter number of standards to be measured for standard curve
Printer	Selects output mode (internal, RS-232, Parallel) (Utility)
Printout Contrast	Allows you to improve visibility of internal printer hardcopy by changing the darkness of the print (Utility)
Protein Factor	Enter factor to calculate Protein concentration (DNA Bio Tests)
Ref. Wavelength	Enter internal reference wavelength value; for each reported measurement, measures analytical wavelength & reference wavelength. Reported measurement = Abs@Analytical WL - Abs@Reference WL
Ref. Wavelength Correction	Turns internal zeroing on or off (**Not** available in Scanning, Multiwavelength, 3-Pt Net and some Bio Tests)
Run Corr.	Initiates the collection of cuvette data for cell correction
Run Standard	Goes to Standards entry screen
Run Test	Goes to the data collection screen (all tests)
Sample Positioner	Select type of Positioner **1 Cell** = no movement (zeros and measures sample in same position (**Not** available in Kinetics and Scanning) **Manual 6** = cell changer moved by buttons (always zeros on position B, then returns to set position to start measurement (**Not** available in Kinetics and Scanning) **Auto 3** = turret auto moved - B, 2, 4 (always zeros on position B, then goes to position 2 to start measurement) (**Not** available in Kinetics and Scanning) **Auto 6** = turret auto moved - B,1,2,3,4,5 (always zeros on position B, then goes to position 1 to start measurement)
Sample Volume	Enters total volume of sample (under Dilution Multiplier in some Bio Tests)
Save Test	Saves all parameters of current test in internal memory for later recall (all tests)

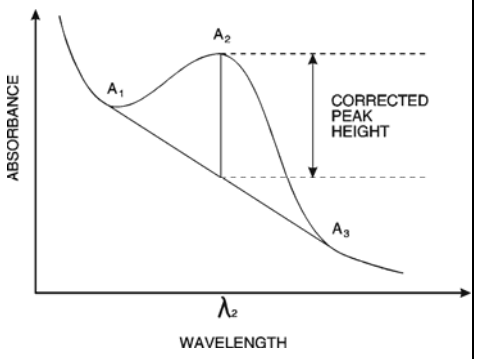
Parameter	Description
Scan speed	Selects speed (nm/min) for a scan – Slow, Medium, Fast (Scanning tests only)
Screen Contrast	Allows you to improve visibility of display by changing contrast between background and text (Utility)
Select Test	Tags highlighted test name with '>' to include test in the SmartStart menu (Utility Stored Tests Directory)
Set Max. X Set Min. X	Set cursor position in graph as the minimum X and the maximum X values for recalculating rate (Kinetics)
Set nms	Allows you to enter and edit wavelength and factor values (Multiwavelength tests)
Set Options	Select factor entry or the baseline to calculate area under the peak in graph (Scanning)
Setup correction	Initiates the procedure to collect the data necessary to correct for absorbance differences between cuvettes (all test types)
Slope	Enter ΔAbs/ΔConcentration value (Standard Curve test type)
Smoothing	Turns data smoothing on and off (Scanning)
Software Revision	Displays version of firmware in instrument (Utility)
SRE tolerance	Enters the acceptable ±Stray radiant energy acceptable
Standard Concentrations	Enters concentration of standards used to generate standard curve for the test
Standby	Selects time since last keystroke or instrument activity; powers down unit to save lamp life (Utility)
Start wavelength	Enter beginning wavelength for a scan (Scanning tests only)
Statistics	Turns stats on or off; if ON, calculates average and Std Dev of results; Statistics registers are cleared when Statistics = OFF and/or when instrument is OFF, and/or when test parameters are changed, and/or when test is saved (or resaved) (in all test types except Kinetics, Scanning, Multiwavelength)

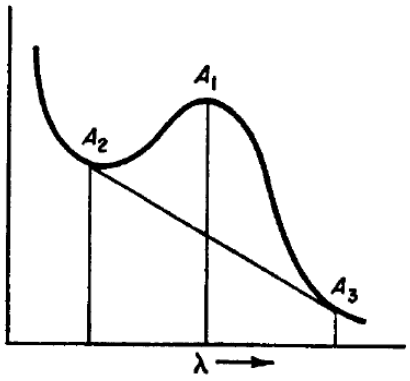
Parameter	Description
Std Concentration	Enters concentration of analyte in standard solution
Stop wavelength	Enter ending wavelength for a scan (Scanning tests only)
Stored Tests Directory	Displays list of tests stored in the instrument (Utility)
Tabular	Displays list of collected data (Kinetics, Scanning)
Test Name	Allows user to enter alphanumeric name (max of 16 characters) for test; name will be included on data printout, and if the test is saved, will be displayed in Utility Test Directory screen (available in all tests)
Tm value	Calculates melting temperature (Oligos tests)
Total Run Time	Enter time from Run initiation to end of test = Delay Time + Interval Times + Measurement Times (Kinetics)
Units	Select or create units labels for results (all stored tests except Abs Ratio, Scanning, Cell Growth)
Unselect Test	Remove '>' tag of highlighted test name to remove test from the SmartStart menu (Utility Stored Tests Directory)
Wavelength	Enter values for the analytical wavelengths

Calculations for Software

Calculation	Calculation(s)	Graphs
Standard Curves		
Partial sums	$SX = \sum x_i$ $SY = \sum y_i$ $SXX = \sum x_i^2$ $SYY = \sum y_i^2$ $SXY = \sum x_i y_i$ $SQX = \sum \left(x_i - \bar{x}\right)^2 = N * SXX - SX^2$ $SQY = \sum \left(y_i - \bar{y}\right)^2 = N * SYY - SY^2$ $SSXY = \sum \left(x_i - \bar{x}\right)\left(y_i - \bar{y}\right)^2 = N * SXY - SX * SY$ where: $x_i =$ concentration of i[th] standard $y_i =$ absorbance of i[th] standard N= number of standards	
Linear regression (general case)	$A = A(c)$ where $A =$ absorbance $c =$ concentration $A(c)$ is defined by an equation of the form $A(C) = a_4 c^4 + a_3 c^3 + a_2 c^2 + a_1 c + a_0$ where $a_0 =$ Y-axis intercept $a_1 \ldots a_4 =$ coefficients The coefficients are computed using the least squares method.	

Calculation	Calculation(s)	Graphs
Linear regression through zero	$A = a_1 * (c)$ where $A =$ absorbance $c =$ concentration $a_1 =$ slope The slope is calculated as $a_1 = SXY / SXX$ This model requires: ▪ Slope is not equal to zero or infinity ▪ At least one standard data point with concentration >0 ▪ The absorbance of the 0 concentration blank = 0A	
Segmented model	The segmented model requires: ▪ Data for at least two standard data points with different concentrations and absorbances ▪ Slopes of all segments must be ascending (positive) ***or*** descending (negative)	
Validity of standard curves	$A(c_1) > A(c_2)$ for all $c_1 > c_2$ or $A(c_1) < A(c_2)$ for all $c_1 > c_2$ where $A =$ absorbance c_1, c_2 concentration	 Valid non-linear standard curve
	If this is not the case, then there will be more than one solution within the specified domain, and the message "Curve cannot be used to determine unknown concentrations – it may produce ambiguous results" will appear when the curve is viewed.	 Invalid non-linear standard curve

Calculation	Calculation(s)	Graphs
Statistics (Linear regression general case)	$\sigma = \sqrt{\frac{\sum\left(y_i - \overline{y_i}\right)^2}{N - n - 1}}$ where $n =$ Degree of polynomial $r = \frac{\lvert SSXY \rvert}{\sqrt{SQX * SQY}}$ Note: The calculation for the correlation coefficient only applies to first-order linear regression curves (first-degree polynomials)	
Linear regression through zero model	$\sigma = \sqrt{\frac{SYY - \left(a_1 * SXY\right)}{N - 1}}$	
Absorbance ratio	$\frac{Abs\lambda_1}{Abs\lambda_2}$ or $\frac{Abs\lambda_1 - Abs_{ref}}{Abs\lambda_2 - Abs_{ref}}$	
Absorbance Difference	Result = $Abs\lambda_1 * factor\ \ 1 - Abs\lambda_2 * factor\ \ 2$ or $\left(Abs\lambda_1 - Abs\lambda_{ref}\right) * factor\ \ 1 - \left(Abs\lambda_2 - Abs\lambda_{ref}\right) * factor\ \ 2$	
3-Point Net	Baseline corrected absorbance = $A_2 - \left(A_3 + \left(\left[A_1 - A_2 \right] * \frac{\lambda_3 - \lambda_2}{\lambda_3 - \lambda_1} \right) \right)$	 3-Point Net Absorbance sample curve

Calculation	Calculation(s)	Graphs
3-Point Net (ASTM E169-04)	Baseline corrected absorbance = $$A_1 - \left(A_3 + \left(\left[A_2 - A_3 \right] * \frac{\lambda_3 - \lambda_1}{\lambda_3 - \lambda_2} \right) \right)$$	

Calculations for Bio Tests Software

Test Name	Calculation(s)	Default Parameters	Displayed Results
DNA/Protein (260, 280)	Dilution Factor (D_1) = $\frac{diluent\ vol + sample\ volume}{sample\ volume}$ DNA concentration = $[(A_1 - A_{ref})f_1 - (A_2 - A_{ref})f_2]D_f$ Protein concentration - $[(A_2 - A_{ref})f_3 - (A_1 - A_{ref})f_4]D_f$ Ratio = $\frac{A_1 - A_{ref}}{A_2 - A_{ref}}$	A_1 = 260 nm A_2 =280 nm (optional) A_{ref} =320 nm (optional) f_1 = 50 f_2 = 1 dil.vol. = 0 smp.vol = 1	µg/mL
DNA (260, 230)	Dilution Factor (D_1) = $\frac{diluent\ vol + sample\ volume}{sample\ volume}$ DNA concentration = $[(A_1 - A_{ref})f_1 - (A_2 - A_{ref})f_2]D_f$ Protein concentration - $[(A_2 - A_{ref})f_3 - (A_1 - A_{ref})f_4]D_f$ Ratio = $\frac{A_1 - A_{ref}}{A_2 - A_{ref}}$	A_1 = 260 nm A_2 =230 nm A_{ref} =320 nm (optional) f_1 = 49.1 f_2 = 3.48 f_3 = 183 f_4 = 75.8 dil.vol. = 0 smp.vol = 1	µg/mL
DNA (260, 280) with Scan	Dilution Factor (D_1) = $\frac{diluent\ vol + sample\ volume}{sample\ volume}$ DNA concentration = $[(A_1 - A_{ref})f_1]D_f$ Protein concentration - $[(A_2 - A_{ref})f_2]D_f$ Ratio = $\frac{A_1 - A_{ref}}{A_2 - A_{ref}}$	Start wavelength = 225 nm Stop wavelength = 325 nm A_1 = 260 nm A_2 =280 nm (optional) A_{ref} =320 nm (optional) f_1 = 50 f_2 = 1 dil.vol. = 0 smp.vol = 0	µg/mL

Test Name	Calculation(s)	Default Parameters	Displayed Results
DNA (260, 230) with scan	Dilution Factor (D_1) = $\frac{diluent\ vol + sample\ volume}{sample\ volume}$ DNA concentration = $[(A_1 - A_{ref})f_1 - (A_2 - A_{ref})f_2]D_f$ Protein concentration - $[(A_2 - A_{ref})f_3 - (A_1 - A_{ref})f_4]D_f$ Ratio = $\frac{A_1 - A_{ref}}{A_2 - A_{ref}}$	A_1 = 260 nm A_2 =230 nm A_{ref} =320 nm (optional) f_1 = 49.1 f_2 = 3.48 f_3 = 183 f_4 = 75.8 dil.vol. = 0 smp.vol = 1	µg/mL
dsDNA	Conc. = $(F \times A_{260})D_f$	260 nm $Factor_{dsDNA} = 50$	µg/mL
ssDNA, RNA	Conc. = $(F \times A_{260})D_f$	260 nm $Factor_{ssDNA}$ or $_{RNA} = 40$	µg/mL
Oligos (entered factor)	Conc. = $(F \times A_{260})D_f$	260 nm $Factor_{oligos} = 33$	µg/mL
Oligos (calc factor)	Conc. = $(F \times A_{260})D_f$ F = factor calculated by Oligo Calculator	260 nm	µg/mL
Bradford -standard	Second order	595 nm Std. Conc. of 0, 200, 400 600, 800, 1000	µg/mL
Bradford -micro	Second order	595 nm Std. Conc. of 0, 20, 40 60, 80, 100	µg/mL
Lowry -standard	Second order	550 nm Std. Conc. of 0, 100, 200 500, 1000, 2000	µg/mL
Lowry -micro	Second order	750 nm Std. Conc. of 0, 20, 50 100, 200, 500	µg/mL
(BCA) -standard	Second order	562 nm Std. Conc. of 0, 0.2, 0.4, 0.6, 0.8, 1.0	µg/mL

Test Name	Calculation(s)	Default Parameters	Displayed Results
(BCA) -micro	Second order	562 nm Std. Conc. of 0, 0.5, 1, 2, 5, 10	µg/mL
Biuret	First order through zero	540 nm Std. Conc. of 0, 2, 4, 6, 8, 10	mg/mL
Direct UV (280)	Conc. = $(F \times A_{280})D_f$	280 nm $Factor_{280} = 1$	mg/mL
Direct UV (205)	Conc. = $(F \times A_{205})D_f$	260 nm $Factor_{205} = 31$	mg/mL
Warburg-Christian	Dilution Factor (D_1) = $\frac{diluent\ vol + sample\ volume}{sample\ volume}$ Protein concentration - $[(A_1)f_1 - (A_2)f_2]D_f$	A_1 = 280 nm A_2 =260 nm f_1 = 1.55 f_2 = 0.76	mg/mL
Cell growth	None	600 nm	Abs

Calculations for Oligo Calculator

Calculation	Entry Parameters	Formula	Displayed Units
# of bases	Repetitive sequence of A, T (or U), G and C	Count total # of bases entered	Length = # of bases
%GC content	Use AT (U) GC sequence entered above	%GC = $\frac{\#\ of\ (G+C)\ bases\ x\ 100}{total\ \#\ of\ AT\ (or\ U)GC}$	Percentage
Molecular weight	# units A, # units T, # units G, # units C # units U	If entry <u>does not</u> include U: MW = (312.2 x A) + (303.2 x T) + (329.2 x G) + (289.2 x C) + 18.02 If entry <u>does</u> include U: MW = (329.2 x A) + (306.2 x U) + (345.2 x G) + (305.2 x C) + 18.02	Molecular weight = x Da/M
Absorptivity ε (260)	# units A, # units T, # units G, # units C # units U	If entry <u>does not</u> include U: ε_{260} = (15,200 x A) + (8,400 x T) + (12,010 x G) + (7,050 x C) If entry <u>does</u> include U: ε_{260} = (15,200 x A) + (9,900 x U) + (12,010 x G) + (7,050 x C)	Extinction coefficient = $M^{-1}cm^{-1}$
Conversion Factor	N/A	Molecular Weight x 10^3 Extinction Coefficient	µg/mL
Calculation of T_m: Oligos up to 20 bases in length	# units A, # units T, # units G, # units C	T_m= 2(A + T) + (G + C)	°C
Calculation of T_m: DNA-DNA hybrids	· # units A, # units T # units G, # units C · M – molarity of cation · Fraction GC = fraction of G and C · %form = %formamide in the sample · L = # of base pairs · P = % mismatching	T_m= 81.5C + 16.6log ((Na+)/(1+0.7(Na+)) + 0.51 (%GC) - 500/L – P – 0.63(%formamide)	°C

Calculation	Entry Parameters	Formula	Displayed Units
Calculation of T_m: DNA-RNA hybrids	· # units A, # units T # units G, # units C · M – molarity of cation · Fraction GC = fraction of G and C · %form = %formamide in the sample · L = # of base pairs · P = % mismatching	T_m= 67°C + 16.6log ((Na+)/ (1+0.7(Na+)) + 0.8 (%GC) - 500/L – P – 0.5(%formamide)	°C
Calculation of T_m: RNA-RNA hybrids	· # units A, # units T # units G, # units C · M – molarity of cation · Fraction GC = fraction of G and C · %form = %formamide in the sample · L = # of base pairs · P = % mismatching	T_m= 78°C + 16.6log ((Na^+)/ (1+0.7(Na^+)) + 0.7 (%GC) - 500/L – P – 0.35(%formamide)	°C
Conversion from	µg/Ml and molecular weight from Oligo (calc factor) test	pmol/µL = $\frac{\mu g / mL \; x \, 1000}{DNA \; Mol. Wt.}$	pmol/µL